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Intratribal Variation among Mature Larvae of Stingless Bees (Apidae: Meliponini) with Descriptions of the Eggs of 11 Species

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ABSTRACT

The abundant members of the Meliponini (stingless bees) are restricted to the tropics and subtropics worldwide. Because past studies of the anatomy of their mature larvae are limited, revealing little anatomical variation in the tribe, the current investigation attempts to examine this topic more thoroughly. Herein we describe the mature larvae of 30 species representing 16 genera of a total world fauna of perhaps 32 genera. Although the larvae of most taxa show little anatomical diversity, two species, *Trigona* (*Duckeola*) *ghilianii* Spinola and *Trigona* (*Frieseomelitta*) *varia* (Lepeletier), have mature larvae that differ remarkably in anatomical structure from those of other known stingless bees, and, further, their individual anatomies suggest a close evolutionary relationship. Larval evidence is presented indicating that *Trichotrigona extranea* (Camargo and Moure) is also related to these two taxa. Because eggs are often included in collections of bee larvae, this study adds descriptions of the eggs of 11 species of the Meliponini not previously characterized as well as a list of those that had been treated earlier.

INTRODUCTION

This study explores data to interpret the evolutionary relationships of taxa assigned to the Meliponini, i.e., the stingless bees, a tribe of perhaps 32 extant generic taxa worldwide (Michener,

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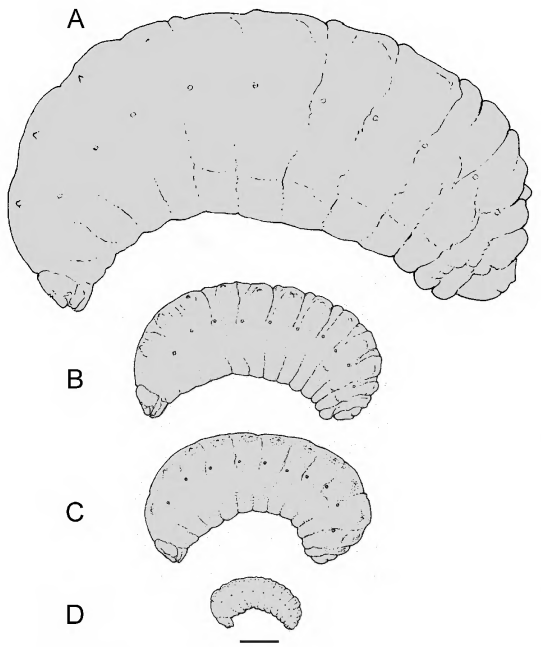


FIGURE 1. Diagrams, all in lateral view and to the same scale, comparing the relative sizes of one of the largest, two of the median group, and one of the smallest of the mature larvae of the Meliponini. **A.** *Melipona* (*Michmelia*) *fallax* Camargo and Pedro. **B.** *Scaptotrigona bipunctata* (Lepeletier). **C.** *Lestrimelitta limao* (Smith). **D.** *Trigonisca mepecheu* Engel and Gonzalez.

2013) within a larger group referred to as the corbiculate bees, comprising honeybees, bumble bees, and euglossine bees in addition to stingless bees. Although most studies of evolutionary relationships of bees are based on adult anatomy and behavior, the current investigation uses the anatomy of mature larvae for these purposes. Examination of mature larvae is undertaken with the hope that it will offer another set of characters to interpret and refine our understanding of these bees and to test the relationships previously proposed using adult anatomy or DNA-based phylogenetic hypotheses (e.g., Michener, 1990; Camargo and Pedro, 1992; Rasmussen and Cameron, 2010).

At the beginning of this study Michener's 2007 detailed classification of the tribe was intended to be followed. However, it soon became apparent that during the following six years he revised his understanding of the Meliponini (Michener, 2013) and concluded that the genus *Trigona* was restricted to the New World, a judgment accepted herein.

In the following survey, although some postdefecating larvae were not available, anatomically fully developed, predefecating specimens or those still defecating were recovered. When a postdefecating form was unavailable, a description of the available defecating or predefecating form was prepared and incorporated into the project. Thus, the term "mature larva" as used here refers to an anatomically fully developed specimen preserved before, during, or after defecation.

Among all collections examined, eggs of many taxa were retrieved. Those of the following had not been described before, so their descriptions and images are included here as an addendum: *Austroplebeia australis* (Friese), *Melipona* (*Eomelipona*) *bicolor* Lepeletier, *Melipona* (*Eomelipona*) *marginata* Lepeletier, *Melipona* (*Melipona*) *quadrifasciata* (Lepeletier), *Melipona* (*Michmelia*) *panamica* Cockerell, *Nannotrigona testaceicornis* (Lepeletier), *Plebeia* (*Plebeia*) *droryana* (Friese), and *Plebeia* (*Plebeia*) "nan2," *Trigona* (*Frieseomelitta*) *varia* (Lepeletier), *Trigona* (*Trigona*) *spinipes* (Fabricius), and *Trigonisca muelleri* (Friese).

Initially presented is a description of the tribe based upon all pre- and postdefecating last larval instars that have been examined by the authors as well as on literature accounts of these forms by Bernadete Lucas de Oliveira, Charles D. Michener, and in several recent papers by

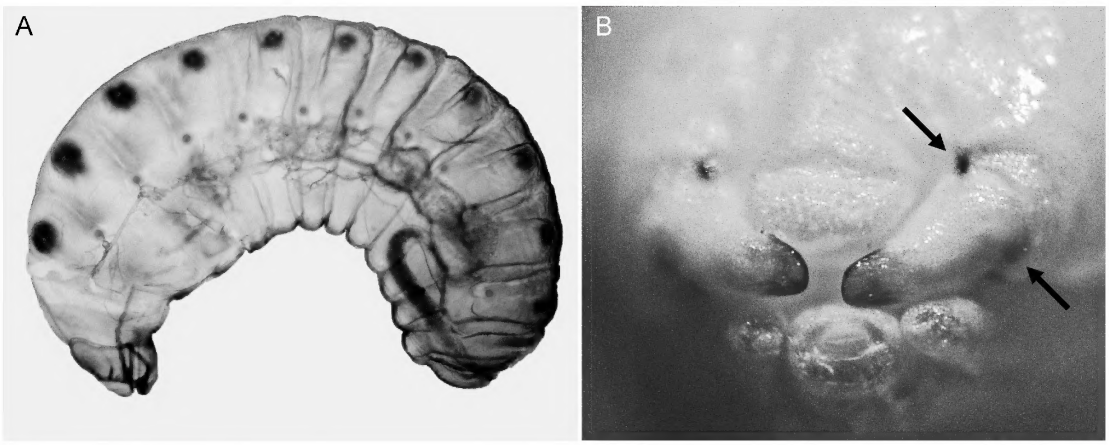


FIGURE 2. **A.** Microphotograph of cleared and lightly stained mature larva of *Paratrigona impunctata* (Ducke), lateral view, showing darkly stained mounds of caudal body annulets relative to lighter stained, membranous integument elsewhere and distribution of ten spiracles (artificially emphasized) on lateral surfaces of left side of cephalic annulets. **B.** Microphotograph of larval head of *Trigona* (*Frieseomelitta*) *varia* (Lepeletier), approximate anterior view showing darkly pigmented anterior articulating points (hinges) on both sides head as well as dark apices of both mandibles (arrows pointing to dorsal and ventral articulations on left side of head). Equally dark posterior points of articulation not clearly visible from this angle, but arrows identify position of anterior and posterior articulation points of left mandible.

J.G. Rozen, Jr. These larvae represent 24 of the 51 extant genera and subgenera currently included in the tribe (Michener, 2013). This is followed by an alphabetically presented series of (1) descriptions of available mature larvae of stingless bees that have not been recorded previously, interspersed with (2) references to descriptions of those that have been so treated. There follows the Addendum, the descriptions and imaging of eggs of 11 species; in addition, table 1 provides an alphabetical reference to all taxa the mature larvae of which are now known with their body lengths where available.

METHODS AND TERMINOLOGY

Our current understanding of larval bee anatomy began with Michener's 1953 "Comparative Morphological and Systematic Studies of Bee Larvae with a Key to the Families of Hymenopterous Larvae." Therein, he not only presented a taxonomic study of the then available larvae of bees but also an overview of larvae of Hymenoptera. The methods that he used are the same as those employed in the current endeavor except his work was carried out before the advent of scanning electron microscopy in entomological laboratories.

For the present study the anatomy of larval specimens required special preparation in two ways:

(1) The larva (with the head capsule partly severed from the body by a small slice into the integument of the first body segment just behind the head) is cleared of internal tissue by being boiled in an aqueous solution of KOH and then transferred to an ethanol solution of Chlorazol Black E for staining. We found it necessary to keep cleared specimens for an hour or more in a moderately dark alcoholic solution of the stain both outside and inside their body cavities.

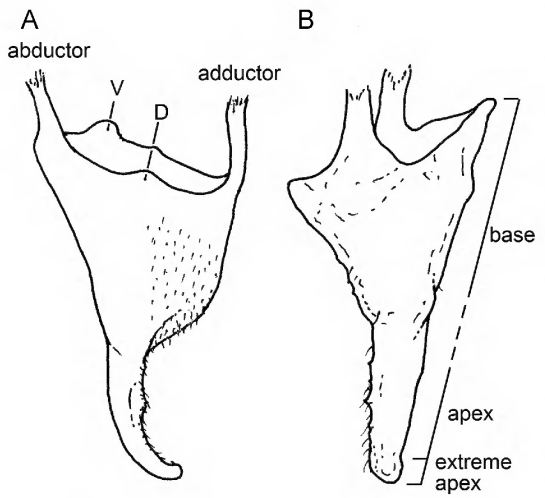


FIGURE 3. Diagrammatic representation of dissected right mandible of *Scaptotrigona depilis* (Moure): **A.** Dorsal view. **B.** Inner view. For explanation, see Methods and Terminology.

After such treatment, while most body integument turns a transparent light blue, the integument of the paired dorsolateral body swellings on caudal annulets becomes an opaque dark blue, as documented (fig. 2A).

(2) In two earlier studies (Rozen and Smith, 2019; Rozen et al., 2019a) minute structures termed multipronged spicules were detected on the apex of the labrum and some surrounding mouthparts of these larvae. Multipronged spicules are sometimes so small that stereoscopic microscopes are unreliable for detecting them, so that SEM imaging is required. For these structures to be examined, two methods may be employed: (1) The larval head is removed from the body, cleared in an aqueous solution of sodium hydroxide, and mounted apically on a copper filament that can be adjust as required by bending prior to SEM examination. (2) The entire uncleared but critical-point dried larva is mounted with its back on the stub so that the front of its head will face the microscope lens after coating.

Table 1 presents data comparing lengths of mature larvae of most of the species. Rozen and Smith (2019) devised a technique for calculating larval lengths of bees by measuring and adding (1) distances between the center of spiracles along one side of the larval body and (2) the two distances separating the extreme ends of the body from centers of their respective closest spiracle. However, that technique had to be modified for these small larvae because the position of spiracles along the side of the body often could not be identified with a stereomicroscope. Instead, measurement of distance between spiracle and extreme ends on camera-lucid diagrams of specimens were recorded and divided by the length of the 1.0 mm scale bar associated with each diagram. So long as each diagram accurately portrays the correct positions of spiracles, such allocations should be reasonably accurate. There remains an unresolved problem in determining information concerning these larvae, i.e., preserved larvae removed from cocoons may have had measurements and shapes that became somewhat distorted by being confined to the shape and dimensions of spheroid cocoons.

Larvae mostly of taxa not previously illustrated in lateral view are presented alphabetically by name in figures 4–30. They are depicted at appropriate magnifications of their body segments to reveal details of their form but irrespective of relative body sizes from one species to the next. All are accompanied by 1 mm scale bars.

For a demonstration of the substantial ranges of body sizes within the tribe, please view the following diagrams, all to the same scale, the largest (fig. 1A) and smallest (fig. 1D) larva studied and that of two representative of the median size (fig. 1B, C).

Mandibles of mature larvae of the Meliponini reveal a good deal of variation requiring description. Using diagrams of the right mandible of *Scaptotrigona depilis* (Moure) as an example (fig. 3A, B), the mandibular **base** is the broad, basal part connected to the head capsule at two articulating points, **D** (dorsal) and **V** (ventral), herein referred to as hinges since they enable the mandible to swing out to open the entrance to the esophagus and swing in for food ingestion and to close the entrance. The basal rim bears two muscle attachments, the **abductor** apodeme and **adductor** apodeme, for the muscles that operate the movement of each mandible. When the abductor muscle contracts, the mandibular apex swings out away from the opening to the esophagus and when the adductor muscles contracts, the mandible swings in, pushing food into the esophagus and afterward, closing the entrance to the digestive tract. The mandibles are connected to the firm head capsule by flexible cuticular conjunctive both externally and adorally, thereby enabling them to easily swing open and close. The mandibular **apex** is the distal, narrowing part of the mandible extending beyond the mandibular base. Although the boundary between the base and apex is imprecisely defined (fig. 3B), the apex is more obviously sclerotized and usually more or less pigmented. With a good many taxa there is a need to recognize the far end of the apex, here termed the **extreme apex**, because of its different shape from the more basal part of the apex and because it often curves abruptly toward the oral opening. The tip of the extreme apex of some taxa is sharply pointed, but in other taxa it is rounded and spoonlike in shape and function. Most but not all mandibles display a dorsal and apical fringe of sharp-pointed spines at the apical end.

As Rozen et al. (2019a: 2), it was anticipated that immatures of some taxa would be described before their valid species names had been established. So that the identity of these specimens could be followed through a series of investigations, the authors proposed "that the generic name of the taxon should be followed by an acronym of 'no available name' and Arabic number, i.e., *Plebeia* 'nan1,' *Plebeia* 'nan2,' etc. These are temporary nomina, without nomenclatorial status." However, when the valid name is established based on the description of the adult following the publication of the acronym, it is important that a clear connection be established between the newly named species and what had previously been published about it before naming.

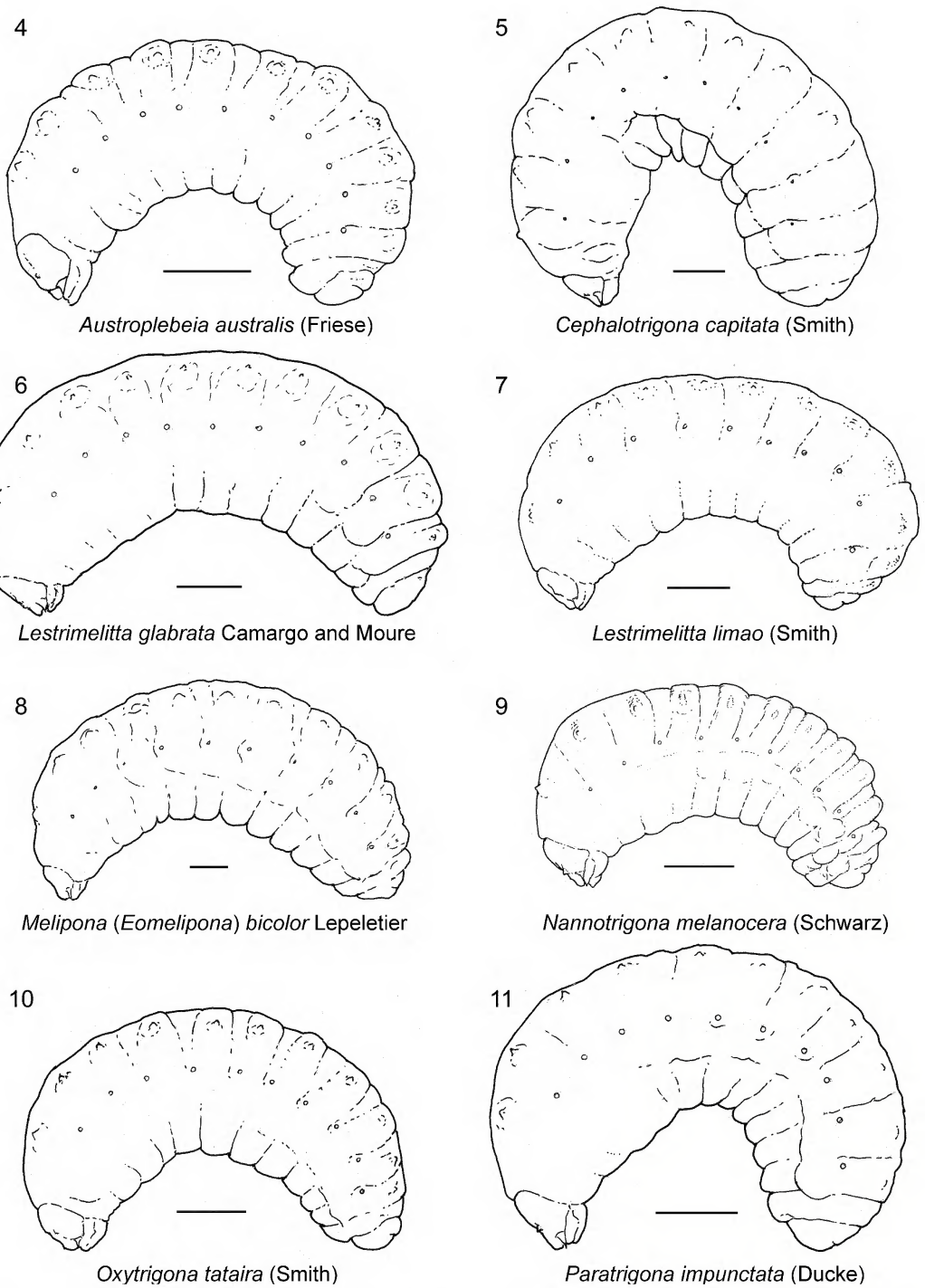
DESCRIPTION OF THE MATURE LARVAL STAGE OF THE TRIBE MELIPONINI

The following description is based upon all species whose mature larvae are now available, including those treated below and species previously described by Rozen et al. (2019a, 2019b) and Rozen and Smith (2019). Although varying considerably in body size, all mature larvae of Meliponini are otherwise remarkably similar except for two known species, namely *Trigona* (*Duckeola*) *ghilianii* (Spinola) and *Trigona* (*Frieseomelitta*) *varia* (Lepeletier), as detailed in the

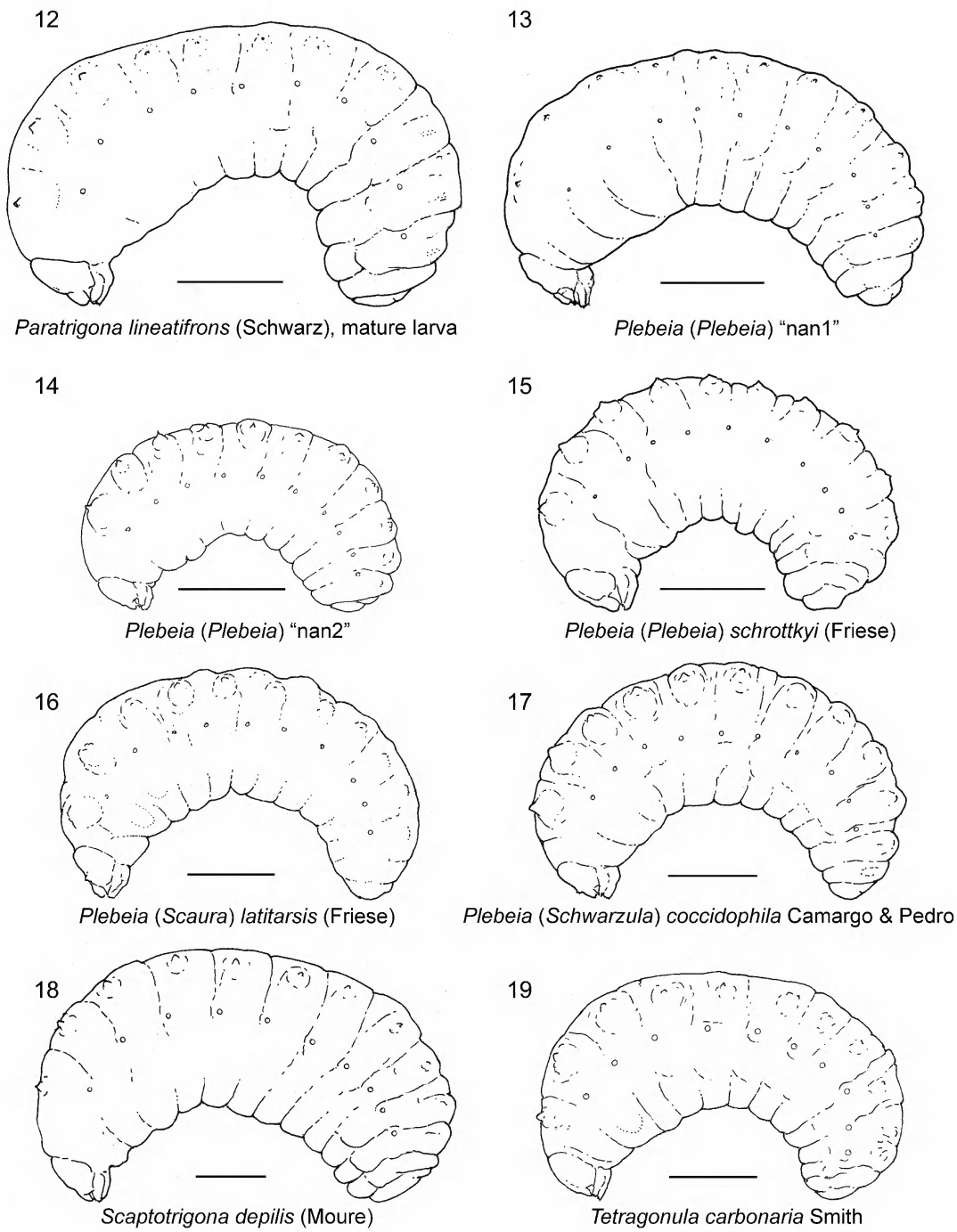
following tribal description and elsewhere herein. A third species, *Trichotrigona extranea* (Camargo and Moure), is also included with these two on the basis of the unpublished thesis of Cristiane Marques Santos (2015), kindly provided by Hugo de Azevedo Werneck. Following the tribal description of the mature larvae of Meliponini is a discussion of some of the characters to convey a fuller understanding of them.

Description: **Head:** Size small (e.g., fig. 9) to moderate (e.g., fig. 4) relative to body size; front of head in lateral profile relatively flat below more or less narrow vertex, so that frons, clypeus, and labrum closely aligned, not projecting in profile beyond labrum, and angling from projecting labiomaxillary region; head capsule consistently broad to very broad in fontal view (e.g., figs. 31A, 42A, 50A); vertex usually not bilobed, but see that of *Trigona* (*Tetragonisca*) *angustula* (Rozen et al, 2019b: fig. 51). Structure of tentorium unknown in postdefecating larvae; posterior tentorial pits normal in position; posterior thickening of head capsule narrow, usually scarcely bending forward medially as seen in dorsal view, but note exceptions in *Trigona* (*Duckeola*) *ghilianii* (fig. 41D) and *Trigona* (*Frieseomelitta*) *varia*; front of head capsule with shallow transverse depression above each antenna and with shallow pair of para-median depressions (both features as well shown in figs. 35A and 45A) between them; area immediately mesad of each parietal band varying from being normally smooth and out-curved (figs. 31A, 45A) to having area depressed and extensively wrinkled (figs. 46A, 50A) because of weak sclerotization (but see additional information below and in Discussion); front of median section of head normally sclerotized, without spicules except in *Trigona* (*Duckeola*) *ghilianii* (fig. 41E) and *Trigona* (*Frieseomelitta*) *varia* (fig. 42B). Parietal bands more or less evident in all species. Antennal papillae large, usually somewhat pointed, thus tuberculate (fig. 35B), but some dome shaped (figs. 46C, 51E); each papilla bearing 3–5 sensilla; papilla surrounded by conspicuous membranous ring (figs. 41E, 42B). Labrum usually more or less broad in frontal view (figs. 34A, 35A, 40A); apical surface of labrum bearing extensive patch of fine, elongate, multipronged spicules (figs. 31B, 37, 41F, 42C) intermixed with sensilla having conspicuous hemispherical bases and mixture of apices some of which are rodlike while others are short and buttonlike; multipronged spicules that are short on upper part of labrum where intermixed with sensilla becoming elongate toward lower labral apex where sensilla are absent.

Figures 4–44. Diagrams in lateral view of available entire mature larvae of Meliponini most of which not illustrated previously, alphabetically arranged by generic/subgeneric names. Although varying greatly in size (as indicated by lengths of 1 mm scale bars on diagrams), bodies are robust, displaying paired, mostly small, dorsolateral tubercles on caudal annulets of first three body (thoracic) segments and on fourth (first abdominal) segment. Paired dorsolateral tubercles of caudal annulets of more posterior segments of most taxa decreasing in size toward posterior end of body. All are postdefecating forms unless stated otherwise.



FIGS. 4–11. Mature larvae of Meliponini. **4.** *Austroplebeia australis* (Friese). **5.** *Cephalotrigona capitata* (Smith). **6.** *Lestrimelitta glabrata* Camargo and Moure. **7.** *Lestrimelitta limao* (Smith). **8.** *Melipona* (*Eomelipona*) *bicolor* Lepeletier. **9.** *Nannotrigona melanocera* (Schwarz). **10.** *Oxytrigona tataira* (Smith). **11.** *Paratrigona impunctata* (Ducke).

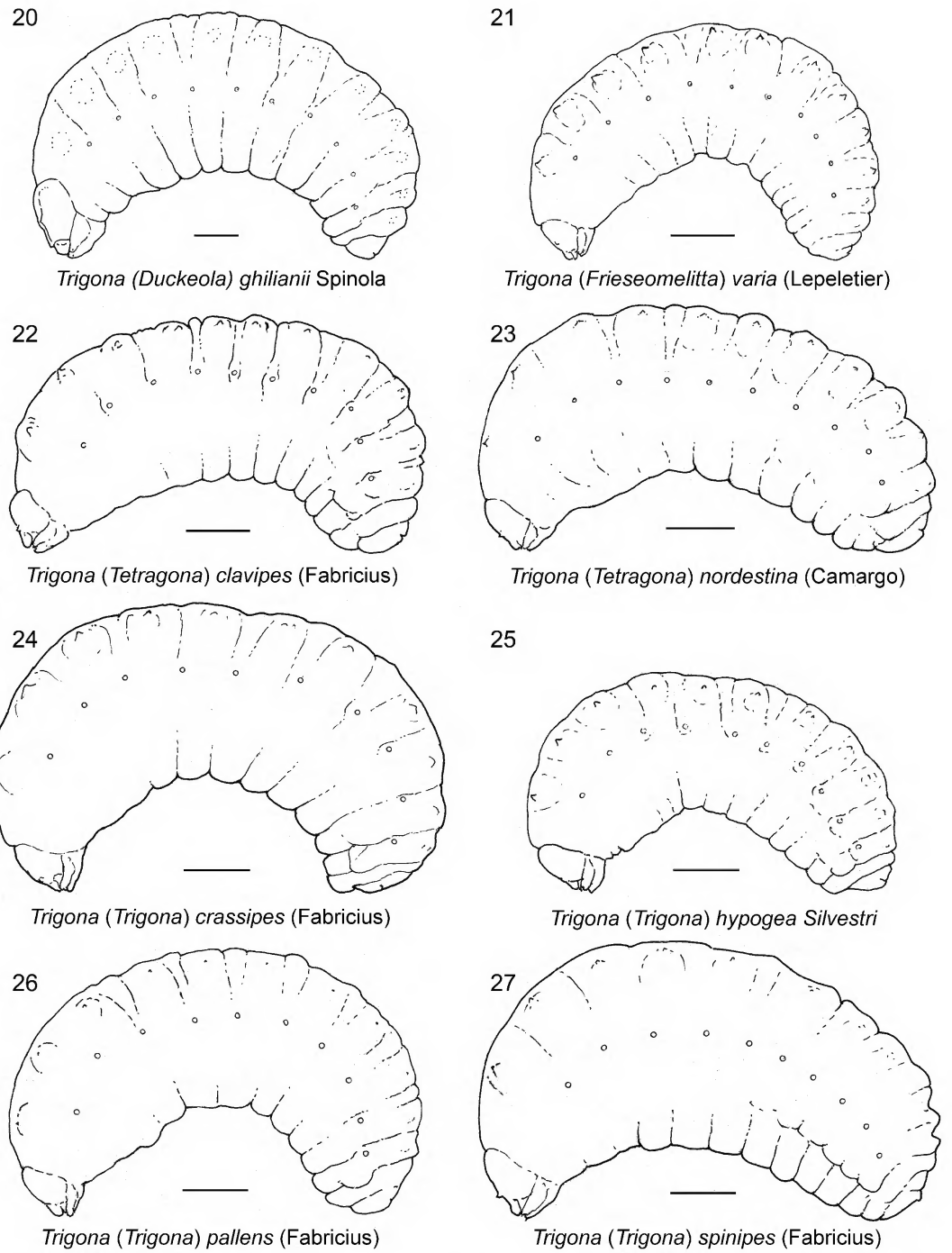


FIGS. 12–19. Mature larvae of Meliponini (*continued*). **12.** *Paratrigona lineatifrons* (Schwarz). **13.** *Plebeia* (*Plebeia*) "nan1." **14.** *Plebeia* (*Plebeia*) "nan2." **15.** *Plebeia* (*Plebeia*) *schrottkyi* (Friese). **16.** *Plebeia* (*Scaura*) *latitarsis* (Friese). **17.** *Plebeia* (*Schwarzula*) *coccidophila* (Camargo and Pedro). **18.** *Scaptotrigona depilis* (Moure). **19.** *Tetragonula carbonaria* Smith.

Mandibular apex of fully developed larva unpigmented to well pigmented, but degree of pigmentation also depending on age of larva; mandible of all species except for *Trigona* (*Frieseomelitta*) *varia* and *Trigona* (*Duckeola*) *ghilianii* (described in detail below in treatment of those species) seen in outer view (e.g., figs. 31B, 35C, 46B, 50B) relatively thick at base, narrowing smoothly to elongate, slender apex; mandibular apex more or less curved adorally and with many taxa having extreme apex abruptly curved adorally, hook shaped; apical concavity narrow, often considerably longer than distance from mandibular base to base of cusp though cuspal base sometimes indistinctly defined; dorsal and apical edges of apical concavity with numerous slender, tapering sharp spines (fig. 48A, B, C); ventral edge without spines or with spines apically, depending on species; cusp more or less projecting, as seen in dorsal or ventral views, depending on species; outer surface of mandibular base somewhat spiculate or not. Length of labial palpus varying from being equal to, to about 2× basal diameter. Surface of maxilla of at least some species ringed by linear series of multipronged spicules suggesting they exert control for directional folding of soft integument (fig. 51C). Hypopharynx spiculate.

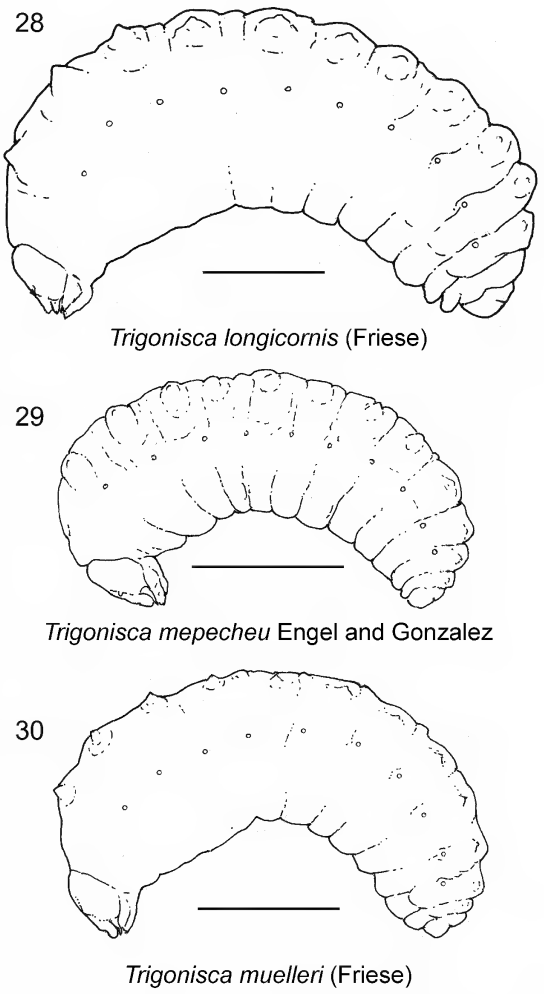
Body: Dorsal integument of body from posterior margin of head to posterior abdominal apex more or less densely covered with very fine, simple (i.e., not multipronged) spicules; this spiculate surface also extending laterally along dorsal surfaces of paired swellings of caudal annulets. Most body segments clearly divided dorsally into cephalic and caudal annulets; caudal annulets on both sides of body tending to be produced as low, transversely oval, dorsolateral swellings that are slightly more sclerotized than rest of integument; these swellings on specimens cleared and stained with Chlorazol Black E (as discussed in Methods, above) acquiring dark blue color; consequently, on cleared and stained specimen (figs. 2A, 39A) most caudal annulets each with sublateral pair of oval elevated, darkly staining, dorsolateral mounds with spiculation on upper surface; these mounds decrease in diameter toward posterior end of body; more anterior caudal annulets each bearing small (sometimes almost obscure), elevated, more or less conical tubercle centered on elevated mound. Thoracic segments externally fused, so that identification of boundaries of annulets difficult to identify, except for paired dorsolateral tubercles on caudal annulet of all three thoracic segments and second and third thoracic segments bearing paired spiracles laterally on cephalic annulets; abdominal segments 1–8 distinctly divided dorsally into cephalic and caudal annulets and each cephalic annulet with lateral paired spiracles; caudal annulets of abdominal segments 7–9 elevated though usually without tubercles; abdominal segments 9 and 10 without spiracles; abdominal segment 10 usually not distinctly divided into cephalic and caudal annulets. All 10 pairs of spiracles moderate in size, peritreme distinct; atrium shallow; atrial wall somewhat annulated; primary tracheal opening a simple rim, smaller than atrial opening; subatrium moderately short; flexure usually collapsed into single narrow tube.

To the extent that they are now known, the mature larvae of the Meliponini are a uniform group in most respects. They possess very robust bodies with head capsules that usually seem disproportionately small compared with their postcephalic bodies. Except for size, their bodies are remarkably similar to one another with most of the 13 body segments each dorsally subdivided into a cephalic and caudal annulet. Each of body segments 2–11 (i.e., thoracic



FIGS. 20–27. Mature larvae of Meliponini (*continued*). **20.** *Trigona* (*Duckeola*) *ghilianii* Spinola. **21.** *Trigona* (*Frieseomelitta*) *varia* (Lepeletier). **22.** *Trigona* (*Tetragona*) *clavipes* (Fabricius). **23.** *Trigona* (*Tetragona*) *nordestina* (Camargo). **24.** *Trigona* (*Trigona*) *crassipes* (Fabricius). **25.** *Trigona* (*Trigona*) *hypogea* Silvestri. **26.** *Trigona* (*Trigona*) *pallens* (Fabricius). **27.** *Trigona* (*Trigona*) *spinipes* (Fabricius).

segments 2 and 3 and abdominal segments 1–8) on all known species bears a pair of spiracles laterally on its cephalic annulet, and all segments bear a pair of dorsolateral swellings on their caudal annulets. The integument of the swellings, especially on the more anterior body segments, though seeming to be membranous, is more sclerotized than that of the posterior segments, best revealed when specimens are stained with Chlorazol E Black. So stained, the darkly colored paired swellings of the first three body segments are large while the swellings of the remaining segments decrease in size toward the posterior end of the body. With most but not all species, each of the dorsolateral caudal annulets of the first four body segments bears a projecting small tubercle, with those of the fourth segment often somewhat smaller than the preceding ones. The more posterior caudal annulets also bear a pair of similar but smaller, less pointed projections that decrease in size toward the posterior end of the abdomen. While these small, paired dorsolateral tubercles on meliponine larvae (as well as most other corbiculate larvae, with the exception of the Apini) have been recognized in the past (e.g., Michener, 2007), their position on the caudal annulets and the expression of the caudal dorsolateral swellings themselves have not been identified in the tribe. The extent of expression of the overall pattern varies from genus to genus but is difficult to use to distinguish taxa because of the subtleties of slight differences in tubercle magnitude among taxa.



FIGS. 28–30. Mature larvae of Meliponini (*continued*). **28.** *Trigonisca longicornis* (Friese). **29.** *Trigonisca mepecheu* Engel and Gonzalez. **30.** *Trigonisca muelleri* (Friese).

Although the thick anterior bodies and small heads give these larvae their robustness, this appearance is further accentuated in postdefecating individuals resulting from development of pupal legs and wings.

The bodies of mature larvae of *Melipona* tend to be large, as indicated by their length (table 1), and the mature larval body of most other genera is shorter and therefore smaller. A visual scanning of the extensive collection of adult specimens at the American Museum of Natural History clearly predicts this conclusion for mature larvae of the entire tribe.

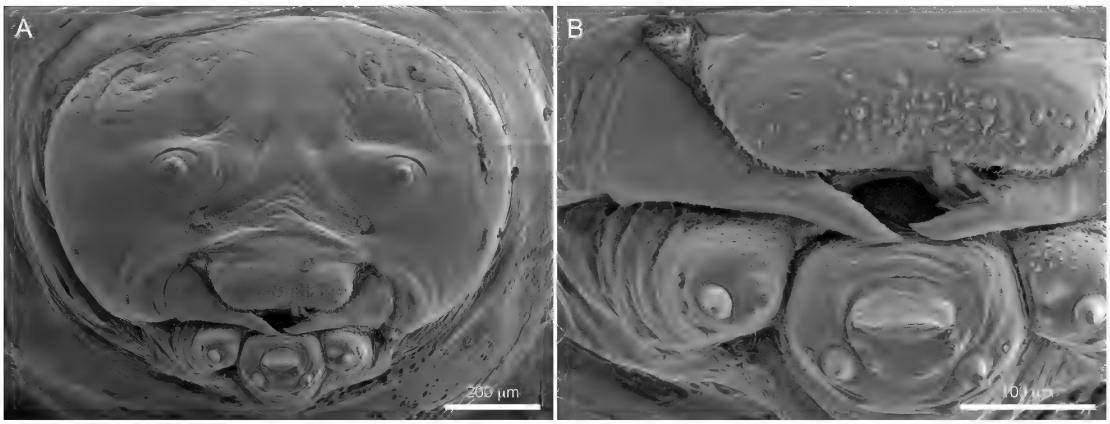


FIG. 31 SEM micrographs of postdefecating larva of *Austroplebeia australis* (Friese): **A.** Head, frontal view. **B.** Close-up of mouthparts.

Head capsules of meliponine mature larvae are broad to very broad as demonstrated herein as well as by previous accounts (Rozen et al., 2019b: figs. 42, 45, 51; Rozen and Smith, 2019: figs. 4, 11; and Rozen et al., 2019a: figs. 7, 12). As pointed out by Rozen et al., 2019a, former authors have sometimes overlooked this feature in that they positioned the illustrated head capsule differently. Also there has been a tendency of displaying one or the other half of the head in frontal view, thereby somehow misjudging the total width of the head capsule compared with its height.

A new feature on heads of some mature bee larvae was recognized when the front of the head capsule was illustrated with a pair of uneven surface features toward the top of the head capsule. Less than a month later, Rozen (in Engel et al., 2019) used SEM micrographs to illustrate (therein fig. 12D, E) the larval head capsule of *Trigonisca mepecheu* with massive wrinkling above each antenna. It was stated (p. 23):

> The extensive wrinkled area above each antenna extending almost to the dorsal edge of the head capsule suggests that the integument there is thin and yielding. In contrast, the smooth surface of the head capsule laterad of the mandibular bases is firm, thickly sclerotized as is the integument surrounding both anterior tentorial pits and the coronal midline of the head capsule. Although there is no internal epistomal ridge mesad to the anterior tentorial pits nor an internal coronal ridge as often found in larval bees…the firm thickened integumental structure of these areas of the head capsule are still present. The two large integumental wrinkled areas laterad of the central coronal thickening occupy areas that are usually smooth curved surfaces on larvae of other bees….

Apparently with these bees there have evolved at least two structural arrangements of head capsule parts, both of which provide a firm base for the energetic functioning of their mandibles.

In the following descriptions of the larvae of various taxa, the degree of distortion of these areas on the front of the head capsule are photographed as observed, but it is doubt-

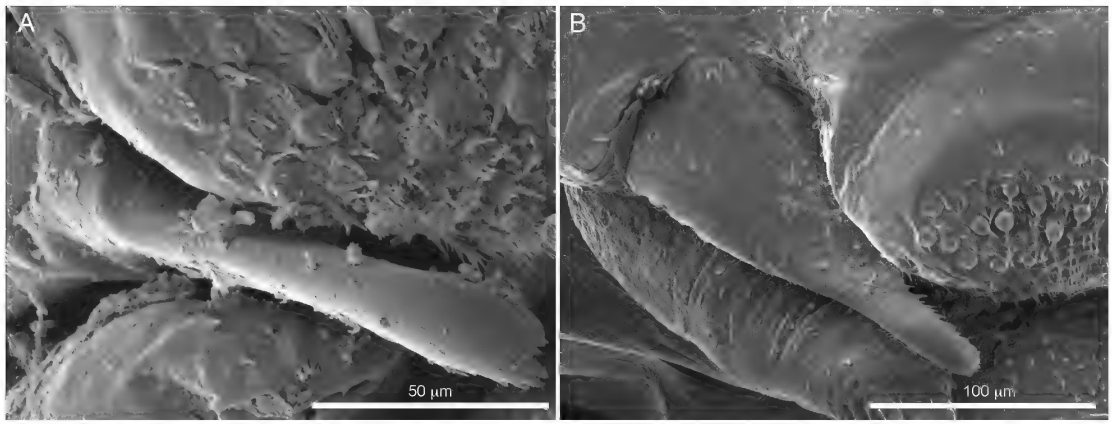


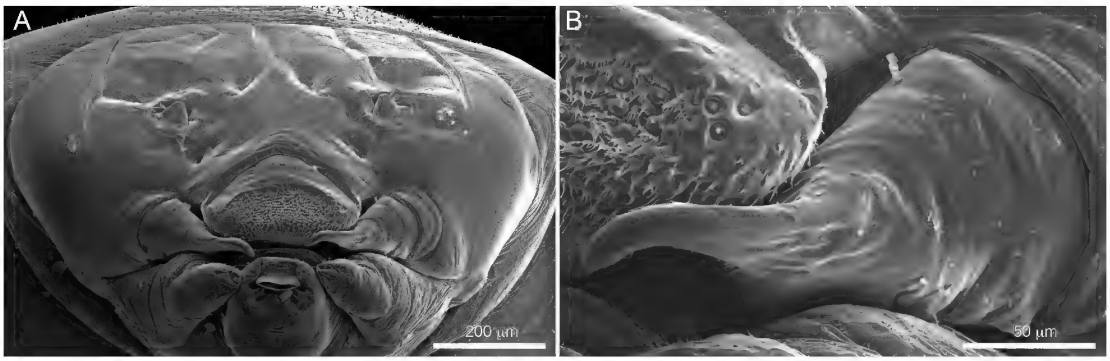
FIG. 32. SEM micrographs of right mandibles of postdefecating larvae of *Lestrimelitta*. **A.** *L. glabrata* Camargo and Moure with apex narrow and extreme apex broadening to become spoon shaped. **B.** *L. limao* (Smith) with apex narrowing gradually all the way toward simple rounded, blunt apex.

ful that they have any diagnostic value. Most likely they are merely random distortions caused by preservation, and, therefore, not described.

The shape of the mandible of mature larvae of most Meliponini detailed in the tribal description is that of a slender structure tapering smoothly from its thick base to its much narrower apex. Its dorsal and apical edges bear sharply pointed spines, and with some species these spines also occur along the ventral apical edge. Contrasting with that configuration was the discovery of a broad mandible first found in *T.* (*Frieseomelitta*) *varia* (figs. 42A, C; 43A–C). Its mandible has a broad and short apical concavity and a short hemispherical mandibular apex, all totally lacking spines. A similar but even more enlarged mandible was later discovered in *T.* (*Duckeola*) *ghilianii* (fig. 41A, B). Both taxa have the dorsal and ventral points of articulation of their mandibles with the head capsule (i.e., mandibular hinges) sclerotized and pigmented (figs. 2B, 43A), unlike in any other known meliponine. Descriptions of these two taxa presented in the following section herein further details contrasting them with the larvae of other tribal members.

Mature larvae of all other meliponines can be identified by their elongate, more strongly tapering mandibles and can be subdivided into three smaller subgroups of taxa on the basis of mandibular morphology, but it is uncertain to what extent, if any, this reflects phylogenetic relationships. In the following lists, generic name alone implies that all species within the genus that have been examined share the feature:

Subgroup 1. Entire mandibular apex tapering from base to extreme apex (e.g., figs. 31B, 48A–C): *Austroplebeia australis* (Friese); *Tetragonula sapiens* (Cockerell), *Melipona, Nannotrigona mirandula* (Cockerell), *Nannotrigona testaceicornis* (Lepeletier), *Paratrigona lineatifrons* (Schwarz), *Plebeia* (*Plebeia*) "nan1," *Scaptotrigona depilis* (Moure), *Trigona* (*Tetragona*) *clavipes* (Fabricius), *Trigona* (*Tetragonisca*) *angustula* Latreille, *Trigona* (*Trigona*) *crassipes* (Fabricius), *Trigonisca longicornis* (Friese), *Trigonisca mepecheu* Engel and Gonzalez.



FIGS. 33. SEM micrographs of postdefecating larva of *Nannotrigona melanocera* (Schwartz). **A.** Head. **B.** Left mandible.

Subgroup 2. Mandibular apex parallel sided before blunt ending (Rozen et al., 2019b, figs. 47, 48): *Plebeia schrottkyi* (Friese), *Nogueirapis mirandula* (Ckll.), *Paratrigona impunctata* (Ducke), and *Partamona musarum* (Ckll.).

Subgroup 3. Mandibular extreme apex expanding in inner or outer views: *Lestrimelitta glabrata* Camargo and Moure (fig. 32A), *Lestrimelitta limao* (Smith) (fig. 32B), *Trigona* (*Tetragona*) *nordestina* (Camargo) (fig. 46).

In addition to postdefecating larvae in these three subgroups, four specimens of predefecating last larval instars of *Cephalotrigona capitata* (Smith) were found to have mandibles intermediate between those of subgroups 2 and 3 in that their mandibular apices expanded faintly. The same could be repeated for a single cleared and stained larva of *Trigona* (*Geotrigona*) *mombuca* Smith almost devoid of pollen in its hind intestine. However, with this larva the slight apical expansion of the mandible is created by the strong array of spines along the lower apical edge of the mandible.

DESCRIPTIONS OF MATURE LARVAE OF INCLUDED SPECIES

Here follows descriptions and usually images of mature larvae of the Meliponini that have not been so treated before but including references to those that have been. Taxa are arranged alphabetically by generic name.

Postdefecating Larva of *Austroplebeia australis* (Friese)

Figures 4, 31A, B

Description: **Head:** Broad in frontal view (fig. 31A); size moderately small relative to body (fig. 4). Antennal papillae hemispherical. Labrum in frontal view short, bearing narrow apical band of sensilla and sparsely mixed with multipronged spicules (fig. 31B).

Mandibular apex pigmented; in outer view (fig. 31B) mandible narrowing to sharply pointed apex, bearing large, sharply pointed spines on dorsal apical edge.

Body: Paired dorsolateral caudal tubercles (fig. 4) short, mound-like projections, decreasing in size on posterior abdominal segments and absent on last three body segments.

Material Studied: 10 last larval instars: Australia, Queensland, Brisbane. 09-XII-2019. E.A.B. Almeida.

FIG. 34. SEM micrograph of head of postdefecating larva of *Oxytrigona tataira* (Smith).

Predefecating Larva of *Cephalotrigona capitata* (Smith)

Figure 5

On dissection the larva of this species selected for illustration and detailed study was found to be a still defecating form. The second larva was considerably smaller but still thought to be the last larval instar.

Description: **Head:** Size small relative to body size (fig. 5). Antennal papilla moderate in size, bearing 5 sensilla. Labrum moderate in size but clearly narrower than distance between antennal papillae.

Mandible narrowing into sclerotized fingerlike projection ending bluntly, scarcely expanding but bearing small spines around apical edge.

Body: Integument spiculate only on dorsal surface. Very low, paired, obtusely pointed tubercles present on caudal annulets of thoracic segments and first abdominal segments and reducing in size on following segments, scarcely evident beyond abdominal segment 5 or 6. Spiracles indistinguishable from those of most other meliponines.

Material Studied: 4 predefecating last larval instars: Brazil, São Paulo, Cajuru. 30-VII-1991. J.M.F. Camargo (Nest 481c).

Postdefecating Larvae of *Lestrimelitta*

Lestrimelitta glabrata Camargo and Moure

Figures 6, 32A

The mature larva of this species of the robber bee genus *Lestrimelitta* is small-headed relative to its moderately large body (fig. 6). The paired dorsolateral body tubercles are small but expressed on most body segments, including abdominal segment 8. The narrow apical mandible with the somewhat spoon-shaped extreme apex (fig. 32A) may be helpful for identification.

Description: **Head:** Size very small relative to body size (fig. 6); vertex not bilobed in frontal view.

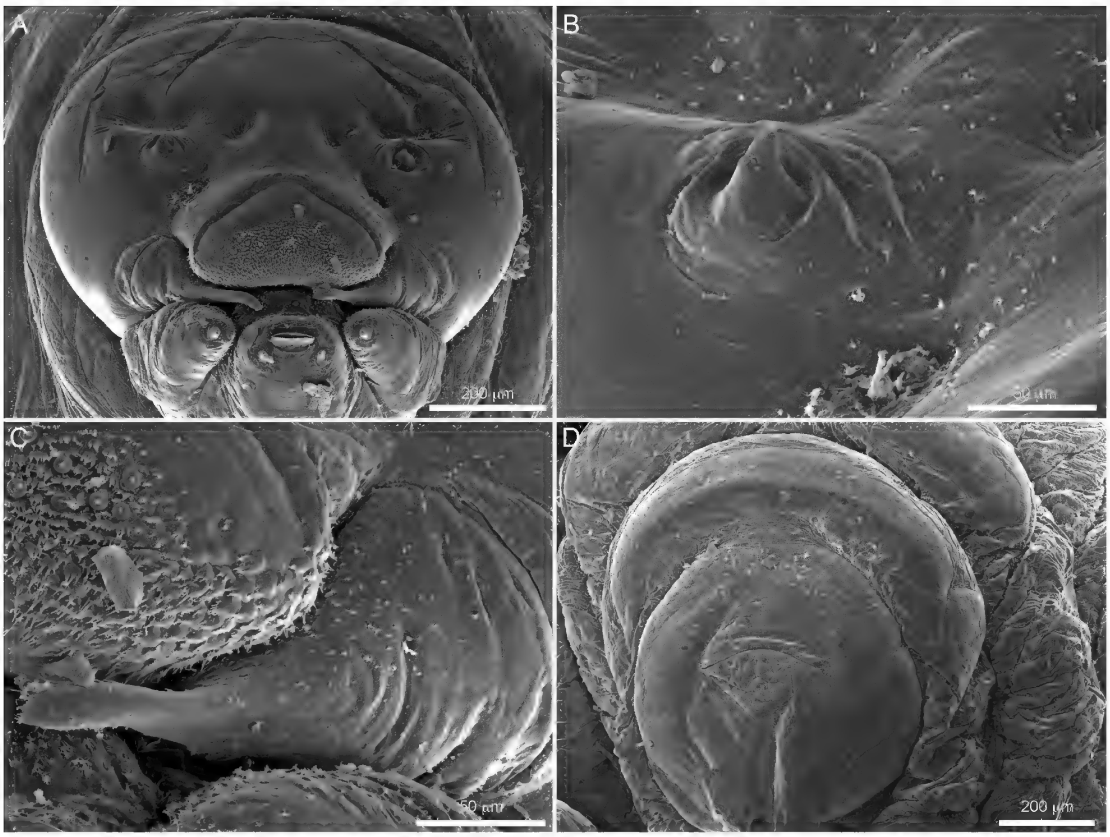


FIG. 35. SEM micrographs of postdefecating larva of *Paratrigona impunctata* (Ducke): **A.** Head. **B.** Right antenna. **C.** Left mandible. **D.** Abdominal segments 10, 9, and 8, posterodorsal view.

Mandibular apex moderately pigmented; mandible seen in outer view (fig. 32A) tapering, subapical section elongate and extremely slender, apex expanding to 1.5× subapical width); apical concavity obscure because of narrowness of mandibular apex; dorsal edge of mandibular apex and of extreme ventral apex with numerous long, slender, sharp spines.

Labiomaxillary region projecting only moderately relative to head capsule in lateral view.

Body: See tribal description.

Material Studied: 10 postdefecating larvae: Brazil, Amazonas, Aruti (rio Negro). 22–23-VIII-1980. J.M.F. Camargo (Nest 234c).

Lestrimelitta limao (Smith)

Figures 7, 32B

The postdefecating larva of this species (fig. 7) is almost identical to that of *Lestrimelitta glabrata* (fig. 7) described above, although the apical breadth of the mandible may be slightly wider and tapers to blunt (non-spoon-shaped) apex (fig. 32B).

Material Studied: 2 postdefecating last larval instars: Brazil, Amazonas, Tapauá. 23-I-1986. J.M.F. Camargo (Nest 343c).

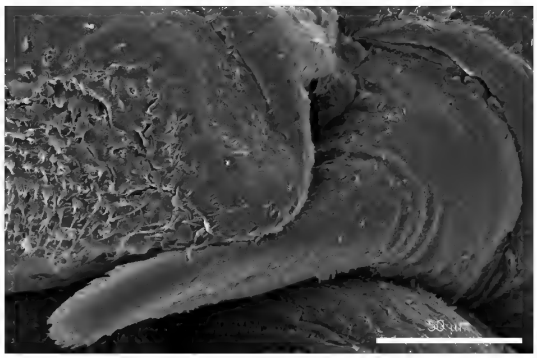


FIG. 36. SEM micrograph of left mandible of postdefecating larva of *Paratrigona lineatifrons* (Schwartz).

Mature Larvae of *Melipona*

All five larval instars of *Melipona nigra schencki* Gribodo were described and illustrated by Lucas de Oliveira (1960). The last larval instars of *Melipona* (*Michmelia*) *fallax* Camargo and Pedro and *Melipona* (*Michmelia*) *trinitatis* Cockerell were recently treated similarly by Rozen and Smith (2019) except now with multipronged spicules identified in figures 13–24 therein for *M. fallax* but also for *Trigona sapiens*. Earlier, Michener (1953) described and illustrated the mature larvae of *M.* (*Eomelipona*) *marginata* Lepeletier, *Melipona* (*Melipona*) *quadrifasciata* (Lepeletier), and *M.* (*Melipona*) *variegatipes* Gribodo.

Melipona (*Eomelipona*) *bicolor* Lepeletier

Figure 8

Except for the strongly pigmented mandibles and small body size (see table 1), the mature larva of this species with its very small head relative to body size (fig. 8) is as described for *M. fallax* (Rozen et al., 2019b: fig. 31).

Material Studied: 1 postdefecating and 2 predefecating larvae: Brazil, São Paulo, Jaguariuna. 27-I-2020. E.A.B. Almeida.

Postdefecating Larvae of *Nannotrigona*

Nannotrigona melanocera (Schwarz)

Figures 9, 33A, B

Description: **Head:** Size moderate relative to body size (fig. 9), broad in frontal view (fig. 34A).

Mandibular apex moderately pigmented; mandible seen in outer view (fig. 33B) slender, narrowing evenly from base to extremely narrow apex; apex bending hook-like toward buccal cavity; dorsal apical edge of mandible with numerous long, slender, sharp spines as seen in inner view but not in outer view (fig. 33B); cuspal area spiculate but not projecting.

Body: Area centered on paired tubercle on caudal annulets of anterior body segments tending to be somewhat transversely elevated (fig. 9); these areas staining readily with

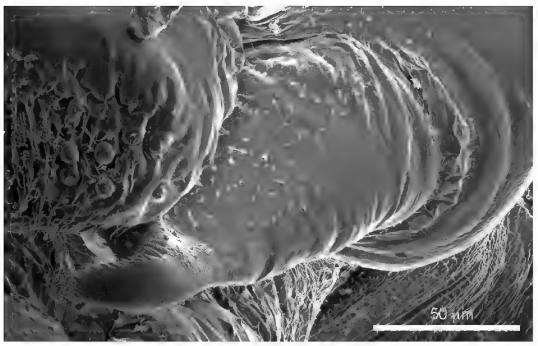


FIG. 37. SEM micrograph of outer surface of left mandible of postdefecating larva of *Plebeia schrottkyi* (Friese).

Chlorazol Black E, so that specimens so treated appear to have dorsal transverse paired treads.

Material Studied: 10+ postdefecating larvae: Brazil, Amazonas, Carauari (rio Juruá). 21-VII-1993. J.M.F. Camargo (Nest 519c).

Nannotrigona testaceicornis (Lepeletier)

Because the larva of this species is nearly identical to that of *N. melanocera,* it is not described or figured herein. One specimen measured 5.9 mm long, somewhat shorter than 6.8, the length of larval *N. melanocera.*

Material Studied: 10 postdefecating larvae: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

Mature Larva of *Nogueirapis mirandula* (Cockerell)

See table 1 and Rozen et al. (2019b) for descriptions of postdefecating larvae and a defecating larva of *Nogueirapis mirandula* from Panamá: Colón Prov., 25 km NE Puerto Pilón on V-22-1986, collected by D. Roubik.

Postdefecating Larva of *Oxytrigona tataira* (Smith)

Figures 10, 34

Description: **Head:** Size compared with body size only moderately small (fig. 10); head capsule wide (fig. 34); vertex somewhat bilobed (fig. 34); antenna tuberculate.

Mandible tapering to thin, elongate, well pigmented, apex; extreme apex slightly swollen, rounded, and bordered by fine spines.

Body: Dorsolateral tubercles on thoracic and basal abdominal segments with short, squat, sharp-pointed projections rising from paired dorsolateral swelling of caudal annulets; these tubercle becoming smaller and more rounded toward posterior end of body.

Material Studied: 3 postdefecating larvae: Brazil, Pará, Tauari. 29-I-1979. J.M.F. Camargo (Nest 165c).

Postdefecating Larvae of *Paratrigona*

As pointed out by Michener (2007), this genus had been divided into two subgenera: *Paratrigona* consisting of a number of species including *P. lineatifrons* and the subgenus

Aparatrigona including *P. impunctata* and one other species. However, Michener did not consider the two subgenera to be sufficiently different to be recognized. Comparisons of the mature larvae of the two representatives described here would seem to support this judgment.

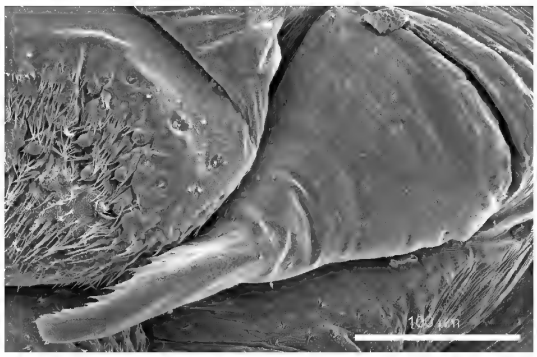


FIG. 38. SEM micrograph of outer surface of left mandible of postdefecating larva of *Scaptotrigona depilis* (Moure).

Paratrigona impunctata (Ducke)

Figures 11, 35A–D

Description: **Head:** Size moderate, small relative to body size (fig. 11). Labrum apically broad, about equal to inter-antennal papillae distance, bearing extensive patch of elongate, multipronged spicules (fig. 35A) medially intermixed above with sensilla having hemispherical bases and often elongate, tapering apices (fig. 35C). Antenna tuberculate (fig. 35B).

Mandibular apex faintly pigmented; mandible seen in outer view (fig. 35C) apically very slender, narrowing evenly from base to apical region, where it becomes parallel sided and without apical expansion; mandibular apex curved adorally and at extreme apex abruptly curved, forming hook-shaped apex; apical concavity extremely narrow; dorsal and apical edges of apical concavity with numerous small, slender, sharp spines faintly visible in (fig. 35C)); ventral edge with spines only apically; in dorsal or ventral views, cuspal surface a continuation of curve of apical concavity.

Body: Thoracic segments and first abdominal segment with small, paired, projecting tubercles on caudal annulets; these tubercles on following abdominal segments scarcely evident; abdominal segment 8 with paired dorsolateral caudal swelling faintly finely spiculate and these swelling almost bandlike on abdominal segments 9 and 10 (fig. 35D).

Material Studied: 4 postdefecating larvae: Brazil, Amazonas, Carauari (rio Juruá). 23-VII-1993. J.M.F. Camargo (Nest 523c).

Paratrigona lineatifrons (Schwarz)

Figures 12, 36

Although somewhat smaller than the postdefecating larva of *P. impunctata* (table 1), the mature larva of *P. lineatifrons* (including its mandible, fig. 36) agrees closely with it.

Material Studied: 8+ mature larvae Brazil, Amazonas, fos do rio Daraá. 02–04-VIII-1980. J.M.F. Camargo (Nest 255c).

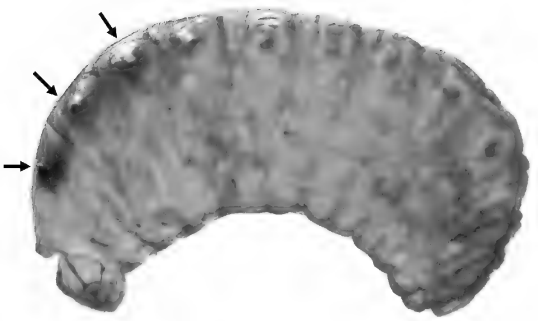


FIG. 39. Microphotographs of entire cleared postdefecating larva of *Scaptotrigona depilis*, lateral view, showing position of small paired dorsolateral tubercles (arrows) on first three body segments.

Postdefecating larvae of *Partamona*

Michener (1953) illustrated and briefly described the larva of *Partamona peckolti* (Friese) as *Trigona* (*Partamona*) *cupira* Smith. However, Rozen et al. (2019b) pointed out "the *Partamona* studied by Michener (1953) could only have been *Partamona peckolti* (Friese); see Moure et al., 2007)."

Partamona cupira (Smith)

Although somewhat shorter (length 7.5 mm), the postdefecating larva of *P. cupira* is otherwise nearly identical to that of *P. musarum* described and illustrated in Rozen et al., 2019b (figs. 44–46), but mislabeled as *P. testacea*. However, one of the last larval instar specimens of *P. cupira* has a small, very dark brown pigmented spot on the apex of each paired dorsolateral tubercle of the thoracic segments and of abdominal segments 1 and 2. These dark brown spots appear dense, opaque, and not the semitranslucent browning of chorion pigmentation. Tubercles on *P. musarum* are without such markings.

Material Studied: 3 postdefecating larvae: Brazil, Minas Gerais, Luislândia. 25-VII-1988. J.M.F. Camargo (Nest 464c).

Partamona musarum (Cockerell)

See Rozen et al. (2019b) for the description of postdefecating larva of *Partamona musarum* (Cockerell) (diagrams therein mislabeled as *P. testacea*). From the original diagram of the larva in lateral view, its length is here reported: 8.8 mm long. Reexamination of the specimen revealed a dense mass of multipronged spicules on the labral apex.

Mature Larvae of *Plebeia*

Following Michener (2007), this genus is here separated into the following subgenera: *Plebeia*, *Scaura*, *Schwarzula*, and *Schwarziana*, with larval representatives of the first three subgenera available for examination.

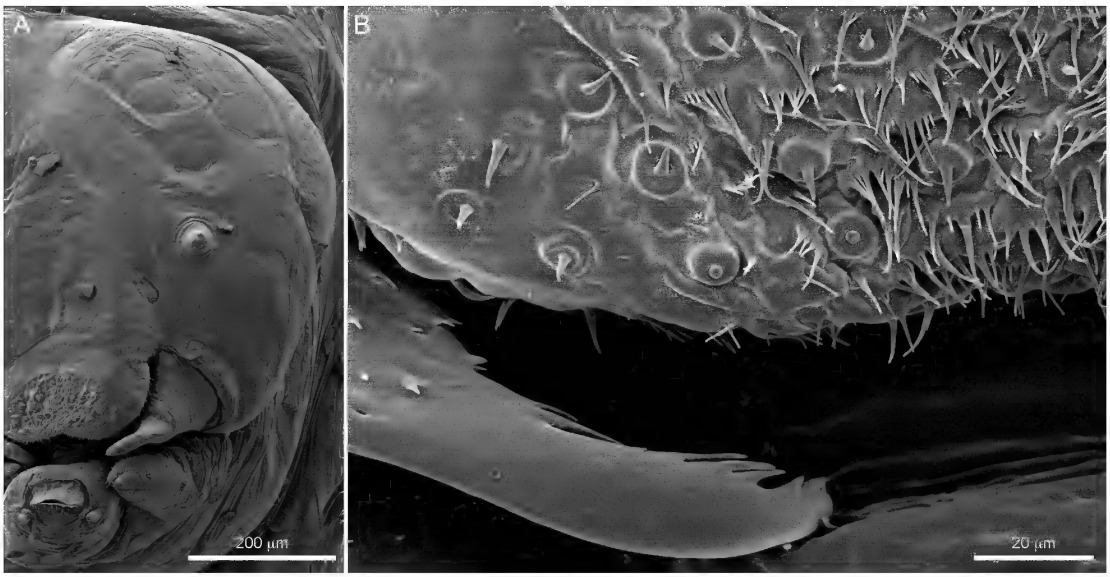


FIG. 40. SEM micrographs of postdefecating larva of *Tetragonula carbonaria* (Smith). **A.** Left side of head, frontal view. **B.** Close-up of spine bearing apex of right mandible below labrum displaying mixture of multipronged spicules and sensilla with hemispherical bases supporting either buttonlike apices or pointed rodlike apices.

Plebeia (*Plebeia*) *droryana* (Friese)

Please see Lucas de Oliveira (1965) for the description and diagram of the predefecating larva of *Plebeia droryana* (Friese). The length of her specimens recorded here in table 1 was derived from her figure 1, which also shows that its head size was moderate compared with body size. This is also true for the 6 postdefecating specimens of the same species listed here.

Material Studied: 6 postdefecating larvae: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

Plebeia (*Plebeia*) "nan1"

Figure 13

Rozen et al. (2019a) described the postdefecating larva of this currently unnamed species of *Plebeia* and in so doing, illustrated it with SEM micrographs (therein, figs. 12–14) of its multipronged spicules. The specimens had been collected on IX-24-2018 by David Roubik from the Yasuni Scientific Field Station in Ecuador as was *Plebeia* "nan2" somewhat more than a year later, in the same tree; see below.

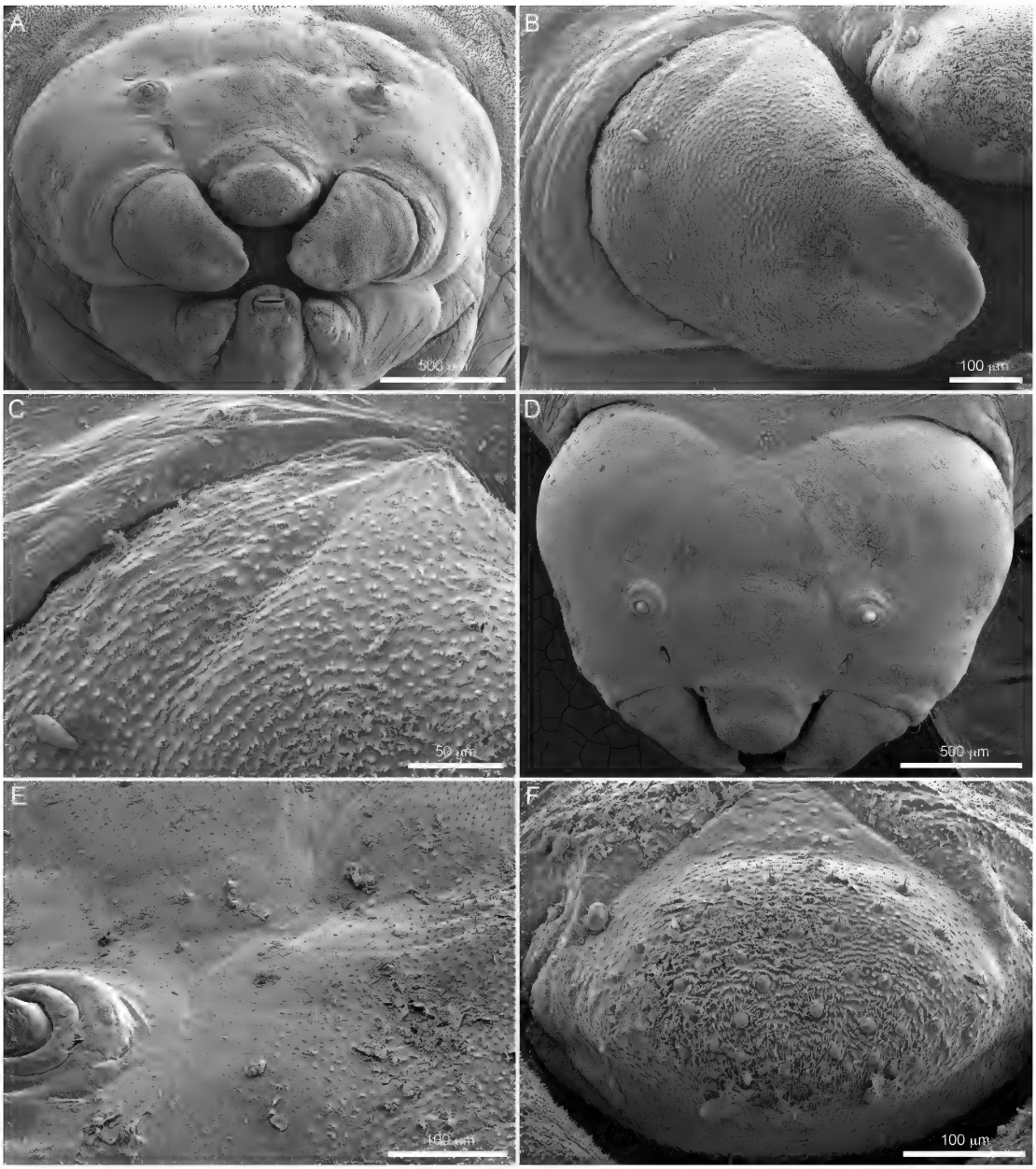


FIG. 41. SEM micrographs of postdefecating larva of *Trigona* (*Duckeola*) *ghilianii* Spinola. **A.** Head, frontal view. **B.** Right mandible. **C.** Close-up of spicules on mandibular surface near dorsal articulation to head. **D.** Dorsal view of head revealing bulging lateral areas of head capsule resulting in V-shaped dorsal posterior margin of head capsule. **E.** Close-up of right antenna and finely spiculate surface along midline of capsule. 41F. Labrum.

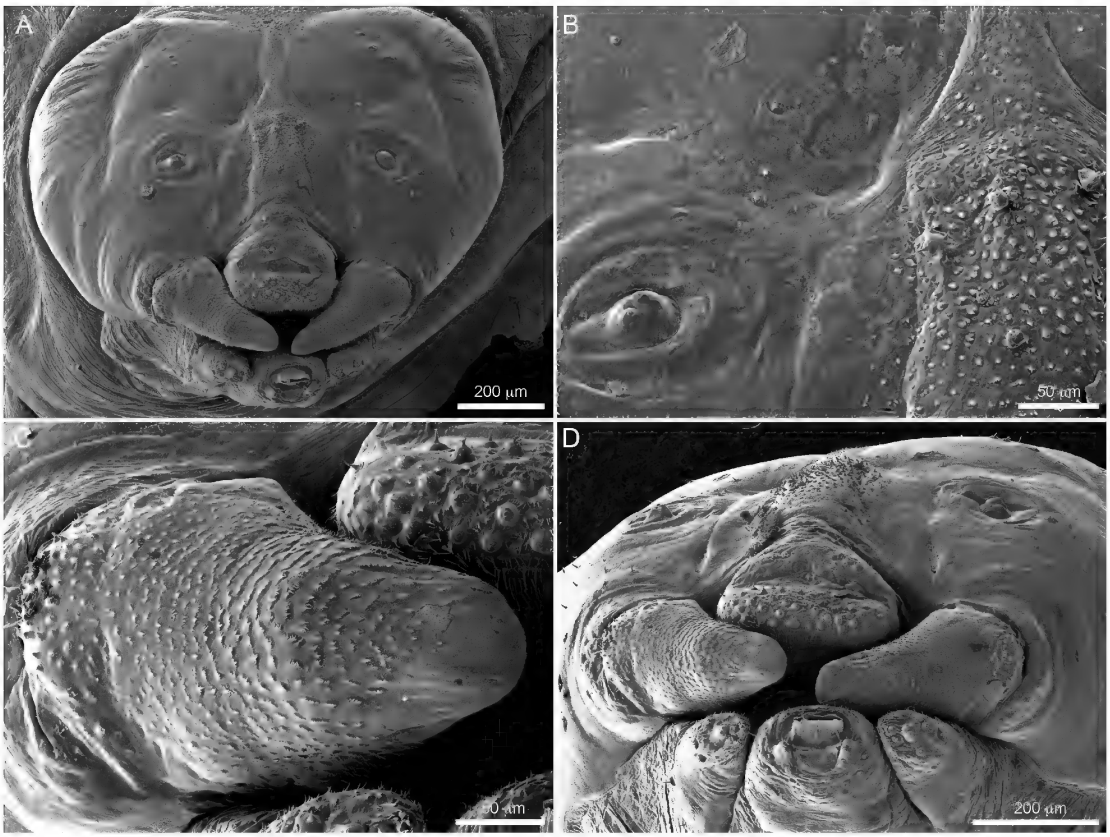


FIG. 42. SEM micrographs of postdefecating larva of *Trigona* (*Frieseomelitta*) *varia* (Lepeletier). **A.** Head, frontal view. **B.** Right antenna and spiculate area along median line of head. **C.** Right mandible with spiculate outer surface and nonspiculate apex. **D.** Head capsule tilted back.

Plebeia (*Plebeia*) "nan2"

Figure 14

The substantially shorter length of this mature larva in contrast to that of *P.* "nan1" (fig. 14) almost certainly indicates that it is as yet another unnamed species of *Plebeia.* Realizing that and recognizing that they had substantially different body sizes, J.G.R. contacted Roubik in an attempt to confirm the understanding of the relationship and nesting behaviors of these two species. His response: "Yes, both in the same tree, and Yasuní Scientific Station continues to be the only place I have found their nests. The second species [*P.* "nan2"] was found by me for the first time about a year ago."

Material Studied: 20+ postdefecating, defecating and predefecating larvae: Ecuador, Francisco de Orellana Prov., Yasuni Scientific Field Station, Pontifica Universidad Católica del Ecuador, -0.8659, -76.3953. Dec. 10, 2019. D. Roubik.

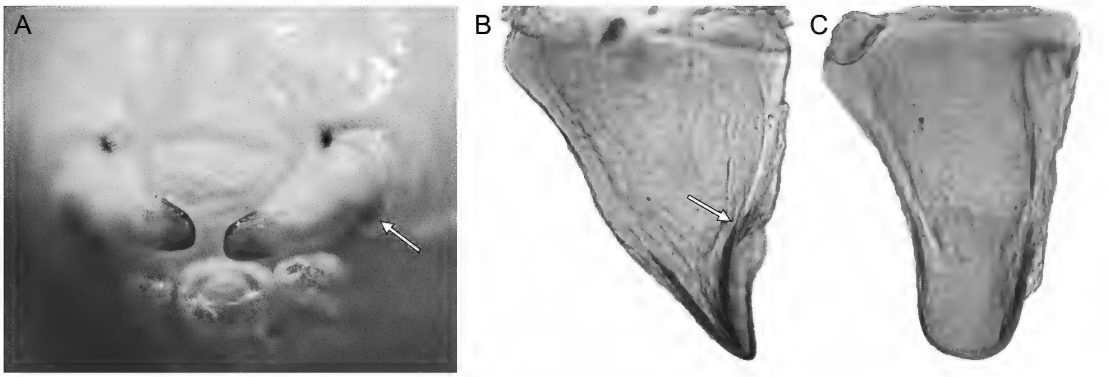


FIG. 43. **A.** Microphotograph of head capsule of *Trigona* (*Frieseomelitta*) *varia* showing darkly pigmented mandibular apices and darkly pigmented dorsal articulations (hinges) and arrow pointing to dimly visible ventral articulation of left mandible. **B, C.** Cleared right mandible, dorsal and inner views.

Plebeia (*Plebeia*) *schrottkyi* (Friese)

Figures 15, 37

Description: **Head:** Size small relative to body size (fig. 15). Antennal papilla tuberculate, moderate in size, bearing 4–5 sensilla. Labrum moderate in size but clearly narrower than distance between antennal papillae; presumably because of poor condition of specimen, multipronged spicules appearing more obscure than in other species relative to somewhat pigmented hemispherical sensilla.

Mandible (fig. 37) tapering from moderately robust base to narrow, pigmented apex that is parallel sided; mandible bearing sharp spines along dorsal apical surface that extend around blunt apical concavity.

Body: As described for *Plebeia* (*Scaura*) *latitarsis* (fig. 16).

Material Studied: 10+ postdefecating larvae: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.B.A. Almeida and D.A. Alves.

Plebeia (*Scaura*) *latitarsis* (Friese)

Figure 16

Of the three larvae available, the one selecting for dissection because of its undistorted anatomy was a predefecating form. Its length of 5.8 mm could only be estimated by measurements derived from the illustration.

Description: **Head:** Size moderately small relative to body size (fig. 16); head not bilobed in frontal view. Vertex in lateral view curving to meet narrow rim of head capsule. Antennal papilla clearly tuberculate. Apical surface of labrum bearing extensive patch of elongate, multipronged spicules.

Mandibular apex even this early stage distinctly pigmented; mandible seen in outer or inner view extremely slender, narrowing evenly from base to very narrow parallel-sided subapical region; extreme apex curving adorally before ending, bearing small, sharply pointed spines on dorsal, ventral, and apical edges.

FIG. 44. SEM micrograph of right mandible of the mature larva of *Trigona* (*Geotrigona*) *mombuca* Smith.

Body: As with other known last larval instar Meliponini, dorsal body surface strongly spiculate on dorsal surface, with spiculation decreasing toward posterior end. Thoracic and most abdominal segments each with clearly defined but small, mostly nonsclerotized tubercle on paired dorsolateral swelling of caudal annulet; on lightly stained specimen these tubercles arising on darkly stained oval surfaces that decrease in size toward posterior end of body.

Material Studied: 3 predefecating and defecating larvae: Brazil, Amazonas, Santa Isabel do Rio Negro. 11-VII-1999. J.M.F. Camargo (Nest 769c).

Plebeia (Schwarzula) *coccidophila* (Camargo and Pedro)

Figure 17

The two smaller larvae identified below appeared in most respects identical to that of *P.* (*Scaura*) *latitarsis* including the same estimated length of 5.8 mm. However, the mandibular apex of *P. coccidophila* is substantial shorter and more robust contrasting with the elongate slender tapering mandibular apex of *P. latitarsis.*

Material Studied: 3 mature larvae (2 smaller specimens including one described containing abundant fecal material restricted to the hind intestine, 1 much larger specimen which may be a predefecating form): Brazil, Amazonas, São Gabriel da Cachoeira. 02 -V3II-1999. J.M.F. Camargo (Nest 791c).

Larvae of *Scaptotrigona*

Scaptotrigona bipunctata (Lepeletier)

The postdefecating larva of this species is nearly identical to that of *S. depilis* except the equally small, paired dorsolateral caudal body tubercles are pigmented.

Material Studied: 7 postdefecating larvae: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

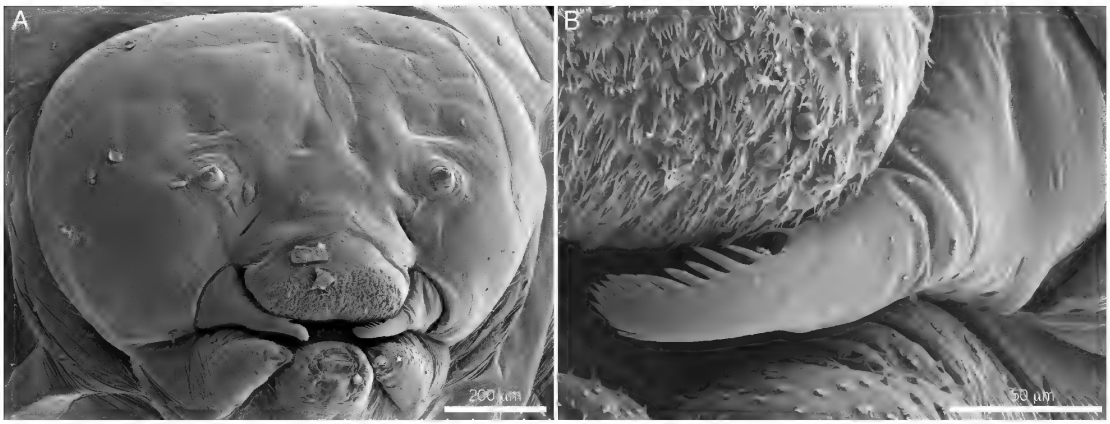


FIG. 45. **A.** SEM micrographs of the head capsule, approximate frontal view, of the mature larva of *Trigona* (*Tetragona*) *clavipes* (Fabricius). **B.** Close-up of left mandible.

Scaptotrigona depilis (Moure)

Figures 3A, B, 18, 39A, B

Description: **Head:** Size small relative to body size (fig. 18).

Mandible tapering from broad base to long slender apex (fig. 39B); apex pigmented; mandible of cleared specimen seen in dorsal, inner, or ventral views (fig. 3A, B) slender, tapering evenly from base to pointed apex; apical concavity narrow, about as long as distance from mandibular base at base of cusp; dorsal edge of apical concavity with numerous long, slender, sharp spines; ventral edge smooth or nearly so; dorsal mandibular surface (fig. 3A) strongly spiculate particularly near basal edge of apical concavity; some spicules also on basal inner surface of concavity.

Body: Anterior body segments with distinct but small, projecting, paired dorsolateral tubercles (fig. 39A), thoracic ones of which tending to be darkly pigmented.

Material Studied: 6+ postdefecating larvae: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.A.B. Almeida and D.A. Alves.

Scaptotrigona "nan1"

The mature larva of this species is similar to that of *S. depilis* and is not illustrated here.

Material Studied: 10+ postdefecating, defecating, and predefecating larvae: Brazil, Amazonas, Acanga, Rio Negro. 06–08-VIII-1980. J.M.F. Camargo (Nest 207c). 15 postdefecating larvae: Brazil, Amazonas, Foz do Rio Curicuriari. 15–21-VII-1980. J.M.F. Camargo (Nest 220c). 6+ postdefecating, defecating, and predefecating larvae: Brazil, Amazonas, Livramento, Rio Negro. 24–26-VII-1980. J.M.F. Camargo.

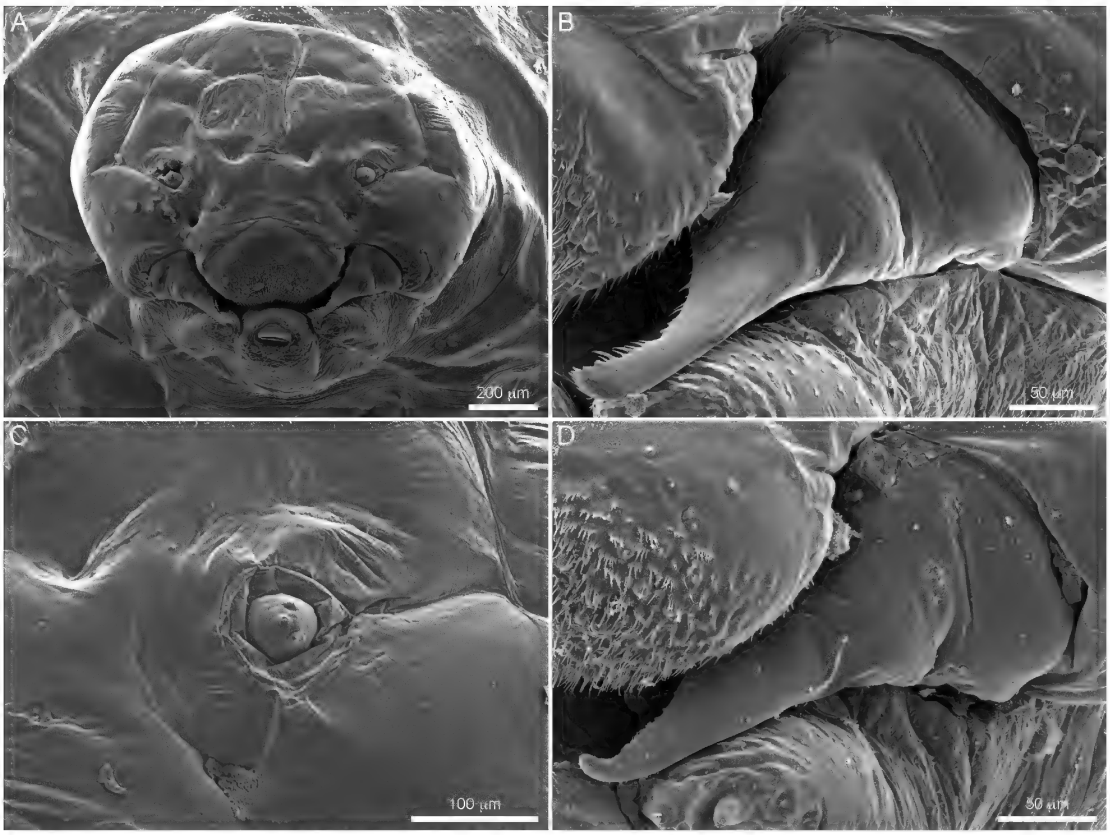


FIG. 46. **A.** SEM micrographs of the head capsule of the mature larva of *Trigona* (*Tetragona*) *nordestina* (Camargo). **B.** Left mandible showing elongate fine spines along dorsal apex. **C.** Left antenna. **D.** Left mandible of another specimen revealing loss of elongate fine spines along dorsal apex.

Scaptotrigona polysticta Moure

The three specimens of this species, though not postdefecating, are nearly identical to the postdefecating larva of *S. depilis.*

Material Studied: 3 defecating or predefecating larvae: Brazil, São Paulo, Jaguariuna. 27-I-2020. E.A.B. Almeida.

Scaptotrigona postica (Latreille)

Lucas (1958) and Lucas de Oliveira (1960) compared the anatomy of the five larval instars of *Scaptotrigona postica* (Latreille) (as *Nannotrigona* (*Scaptotrigona*) *postica* (Latreille)) with those of *Melipona* (*Eomelipona*) *bicolor schencki* Gribodo (as *Melipona nigra schencki* Gribodo). The detailed illustrations of the heads and mandibles of the larval instars in the 1958 paper are admirable.

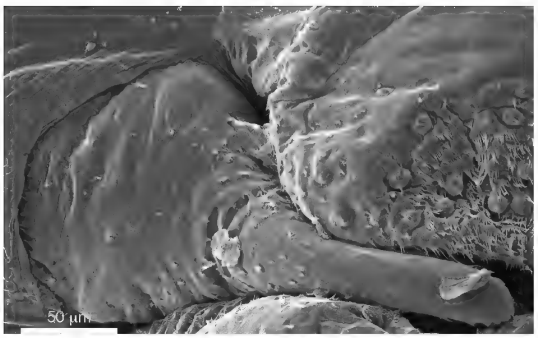


FIG. 47. SEM micrograph of right mandible of *Trigona* (*Trigona*) *crassipes* (Fabricius).

Both preserved larval specimens available for examination were defecating or predefecating forms. After being cleared, one measured 8.1 mm, obviously not comparable to measurements based on noncleared, postdefecating forms. The paired dorsolateral tubercles of the three thoracic segments are small but evident, those of the first abdominal segment are smaller, and those of the following abdominal segments are scarcely evident. Mandibles are tapering with a narrow, darkly pigmented, sharply pointed apex bearing sharp spines.

Material Studied: 2 mature larvae: Brazil, Tocantins, Kraolandia. 20-I-1993. J.M.F. Camargo (Nest 495c).

Mature Larva of *Trichotrigona extranea* Camargo and Moure

In the thesis of Cristiane Marques Santos (2015) made available through the efforts of Hugo de Azevedo Werneck there is a description of the last larval instar of *Trichotrigona extranea* (Camargo and Moure) included in a massive array of descriptions of other larval Meliponini. The descriptions are accompanied by extensive microphotographs (**identified herein with boldface numbers that refer to the thesis**) of head capsules and mouthparts especially including mandibles. The following description of the mandible of *Trichotrigona extranea* based on her dorsal microphotograph (**5d**) of the right mandible and inner microphotograph of the left mandible, which are accompanied by comparative micrographs of mandibles of both *Trigona* (*Duckeola*) *ghilianii* (**5c**) and *T.* (*Frieseomelitta*) *varia* (**5e**). The mandible of these three taxa have apices that are not elongate compared with their basal width. Furthermore, their extreme apices do not bear a fringe of sharp-pointed spines. The outer mandibular surfaces of *T.* (*D.*) *ghilianii* (**5c**) and *T.* (*F.*) *varia* (**5e**) certainly exhibit a dense covering of multipronged spicules, although such surfaces are better displayed herein (figs. 41B, C, 42C). Mandibles of other meliponine larvae exhibit at most only limited accumulations of multipronged spicules restricted to the mandibular base. The mandibles of *Trichotrigona extranea* (**5d**) clearly exhibit vestiture, but the microphotographs do not clearly reveal its composition. Nevertheless, evidence of an extensive display of multipronged spicules is revealed (**3b**) on the labrum and on the outer mandibular surface, lower right corner (Santos, 2015).

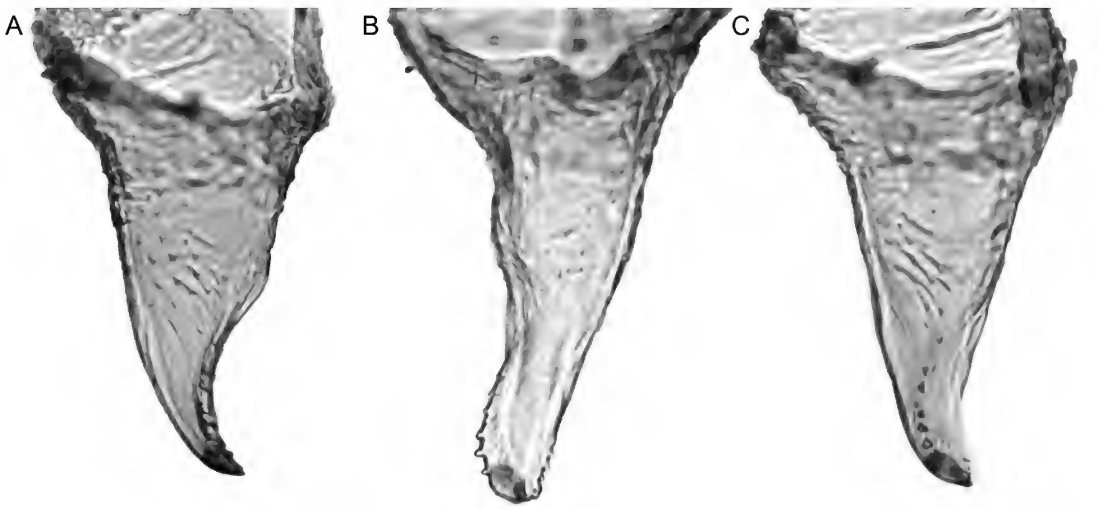


FIG. 48. Microphotographs of right mandible of *Trigona crassipes*, in **A,** dorsal and **B,** inner views, and **C,** partly rotated to exhibit apical concavity.

Postdefecating Larvae of *Trigona*

Postdefecating Larva of *Trigona* (*Duckeola*) *ghilianii* (Spinola)

Figures 20, 41A–F

The huge mandibles and complete lack of body tubercles distinguish the larva of this species from those of all other stingless bees whose larvae have been examined for this study. However, the massive mandibular base and broad apex with a multipronged spiculate outer surface also occurs in *T.* (*Frieseomelitta*) *varia.* Furthermore, the narrow maximum width of the labrum relative to either maximum head or intra-antennal distance of these two taxa also separate them from other meliponine larvae examined for this study. All other stingless bees examined herein have broader labra and more slender mandibles with narrow, slender apices and outer surfaces mostly devoid of multipronged spicules. Although *T.* (*Duckeola*) *ghilianii* and *T.* (*Frieseomelitta*) *varia* share enlarged mandibles externally covered with multipronged spicules, mandibles of *T.*(*D.*) *ghilianii* (fig. 41A, B) are much more enlarged than those of *T.*(*F.*) *varia* (fig. 42A, C) as is the internal structural framework of the head capsule required to support them. Note also the differences in the relative sizes of the labral widths compared with that of the mandibular bases.

Description: **Head:** Size moderate relative to body size (fig. 20); vertex bilobed in frontal view, but, when viewed dorsally, posterior thickening of head capsule strongly V-shaped (fig. 41D) because parietals strongly developed laterally, presumably to accommodate strong

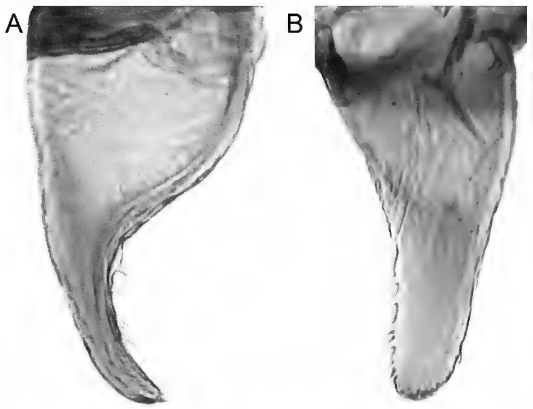


FIG. 49. Microphotograph of right mandible of *Trigona spinipes,* **A,** dorsal and **B,** inner views.

mandibular musculature and resulting in extensive width of head capsule; interantennal integument above clypeus weakly spiculate in limited areas (fig. 41E) but far less conspicuously so than in *T.* (*Frieseomelitta*) *varia* (fig. 42B); these spicules both simple and multipronged. Head integument thick, especially in areas supporting attachment of mandibles (best observed in cleared head capsule. Labrum (fig. 41A, D) small relative to width of head, with mixture of very short multipronged spicules and conspicuous sensilla on hemispherical bases (fig. 41F).

Mandibular base massively swollen (fig. 41A–C); mandibular apex strongly pigmented, appearing as small swelling without spicules at apex of huge mandibular base (fig. 41B); apical concavity small, restricted to inner surface of small apex; front and rear articulation points of mandible (hinges) darkly pigmented, as are these points on head capsule (all as in *T.*(*F.*) *varia*, fig. 43A); much of outer surface of mandible covered by extremely fine spicules, some or perhaps most of which are multipronged (fig. 41B, C). Labiomaxillary region with apical elements small relative to huge size of mandibles (fig. 41A).

Body: Integument strongly spiculate only dorsally; paired oval dorsolateral areas of caudal annulets evident on most segments, but none bearing tubercles (fig. 20).

Material Studied: 4 mature larvae: Brazil, Amazonas, Gavião. 31-VII-1999. J.M.F. Camargo (Nest 833c).

Postdefecating Larva of *Trigona* (*Frieseomelitta*) *varia* (Lepeletier)

Figures 21, 42A–D, 43A–C

The extensive similarities of the larva of this species to those of *T.* (*Duckeola*) *ghilianii* are pointed out above in the initial part of the treatment of that species.

Description: **Head:** Head size small relative to body; vertex not bilobed in frontal view (fig. 42A); parietals swollen outward and backward, presumably accommodating attachment of massive mandibular muscles; posterior margin of each parietal in dorsal view bending forward toward median line as if they would intersect at 55° angle, but posterior margins ending before meeting because midline of head capsule membranous (non-sclerotized) and spiculate with both simple and weakly multipronged. Apical surface of labrum relatively narrow, displaying extensive number of hemispherical sensilla intermixed with multipronged spicules (fig. 42A, B); sensilla arranged mostly along lower edge of labrum, consisting of those with apical buttons and others with apical tapering rods (see Comments, below).

Mandibular apex strongly pigmented (43A); mandible seen in inner or outer views (figs. 42A, D, 43C) massive, robust, evenly tapering to dark brown, broad, hemispherical apex in sharp contrast to elongate, narrowed mandibular apex of most other known mature meliponine larvae (cf. tribal description); most of outer surface of mandible densely covered with multipronged spicules directed toward mandibular apex (fig. 42C); spicules fading out at apex; apical concavity (fig. 43B) short and broad; all surfaces of mandible without spines or setae; anterior and posterior corners of mandibular articulations (hinges) with head capsule darkly pigmented perhaps suggesting enhanced strength required (see Comments, below).

Other mouthparts as described for tribe.

Body: Dorsal integument of thorax spiculate as described for tribe; tubercles and ridges of body as described for tribe, but extent of projection of dorsolateral caudal surfaces and tubercles tending to be somewhat greater than these surfaces in other known tribal taxa. Each thoracic segment with pair of small but distinct, elevated, dorsolateral conical tubercles on caudal annulets; each tubercle mounted on low, transversely oblong mound; abdominal segment 1 to 6 or 7 each also with paired, similarly mounted tubercles, but these tubercles less acute and becoming sequentially smaller toward posterior end of body; abdominal segment 8 with small paired transverse oblong partly raised areas spiculate but lacking tubercles; dorsal surfaces of segments 9 and 10 each with darkly staining elevated bar. Spiracles moderately large relative to body size, as described for tribe.

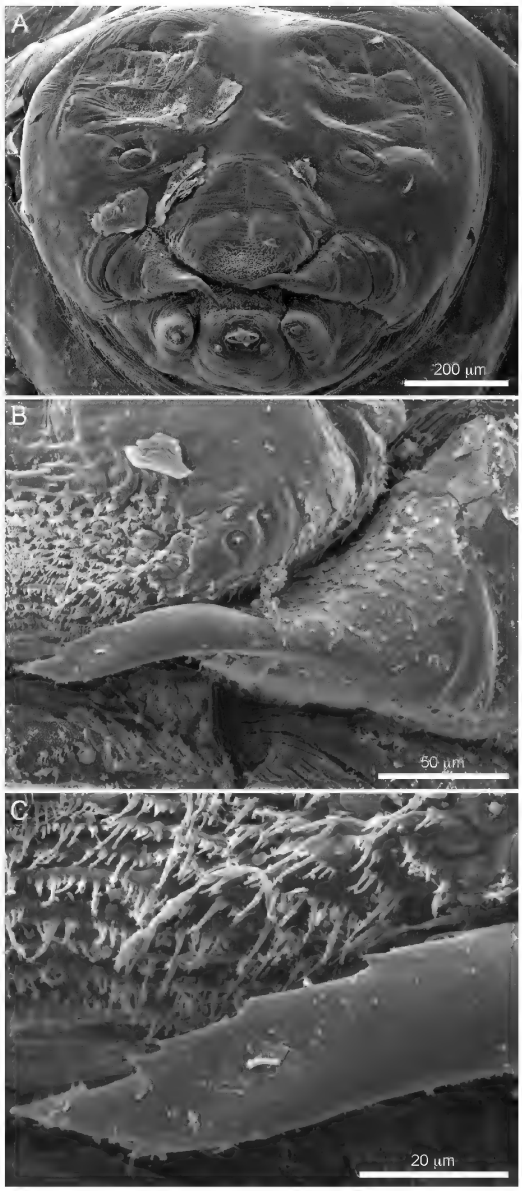


FIG. 50. SEM Micrographs of the head of the post-defecating larva of *Trigonisca longicornis* (Friese). **A.** Head, fontal view. **B.** Left mandible. **C.** Close-up mandibular apex.

Comments: The broad outer surface of the mandible with a dense covering of apically pointed multipronged spicules (fig. 42A, C, D, a feature shared only with *T.* (*Duckeola*) *ghilianii* (see above), suggests the spicules have an important function: when they may act to modify the boundary layer between the mandible surface and the highly viscous fluid such as nectar and in doing so reduce the drag on the moving mandibles during feeding. Thus, they may enable the mandible in the viscous

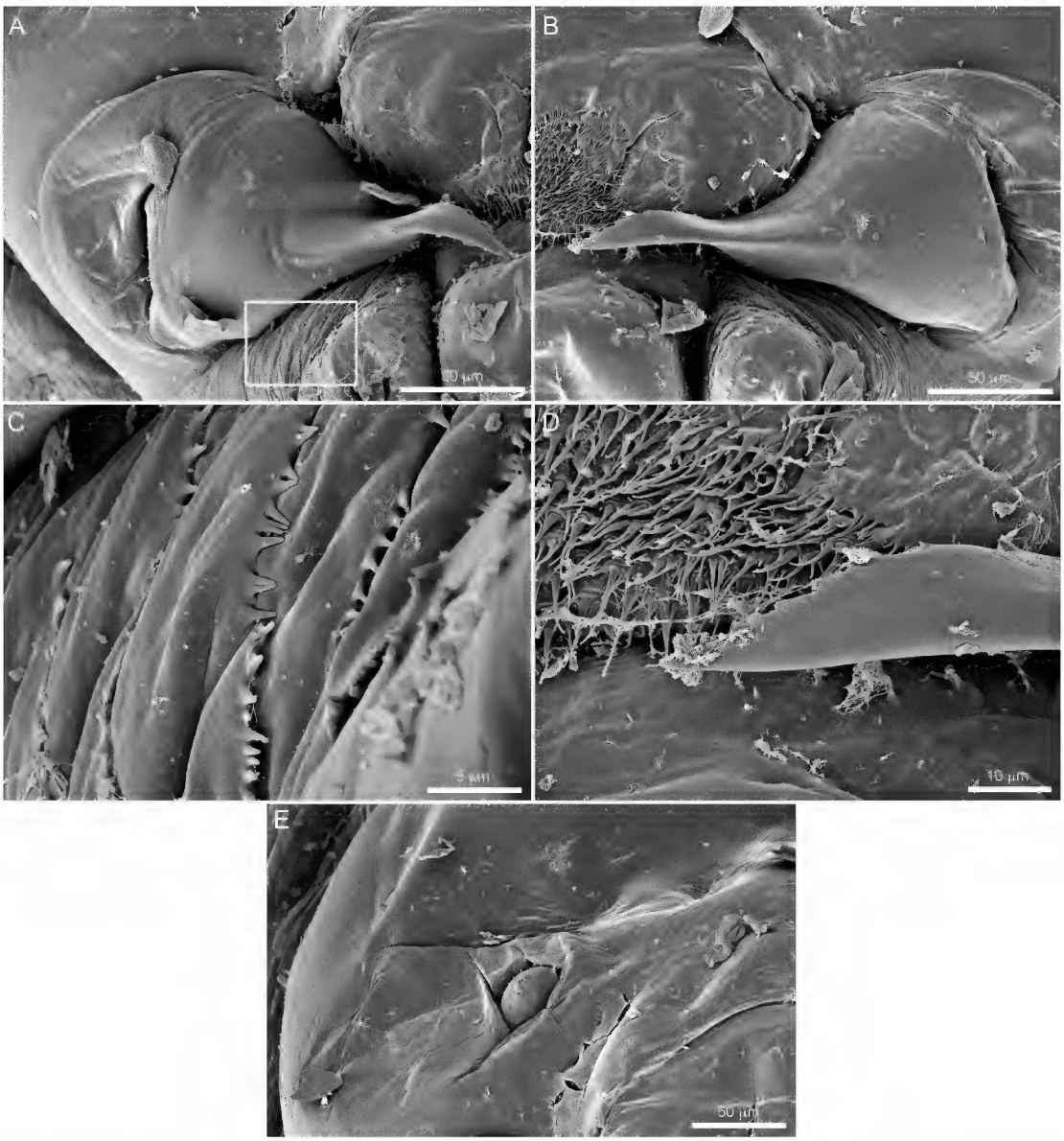


FIG. 51. SEM micrographs of head of immature larva (perhaps fourth instar) of *Trigonisca muelleri* (Friese) (in absence of suitable image of mature larva) revealing large dome-shaped antenna (fig. 51E) and mandibles that taper strongly from broad base to pointed apex (fig. 51A, B). 51C. Close-up of outer surface of the right maxilla identified by rectangle in figure 51A revealing rows of multipronged spicules and providing insight how these rows on flexible integument control bending of maxillary apex while not limiting extension or contraction of apex. 51D. Close-up of apex of left mandible showing sharp tip and jagged dorsal edge.

nectar to open quickly so that viscous food (nectar and pollen) can be ingested easily with the closing of the mandible. The broad outer surface of the mandible becomes more slippery because of some physical action applied to the boundary layer by the many projecting prongs, all of which have a tapering shape. It is unclear what the physical action might be, but it should be noted that the strong narrowing of the mandibular apices of almost all the other studied meliponine larvae reported here presumably enhance the closing and opening of their mandibles through reduction of the burden of pushing through viscous nectar.

The distinctive larval anatomy of *T.* (*Frieseomelitta*) *varia* raises interesting questions. Why are the labral sensilla on hemispherical elevations (fig. 42A, C, D) not only on this species but all known Meliponini? Might their elevation enable them to detect sense stimuli that would otherwise be entrapped by a surface layer of nectar created by the multipronged spicules?

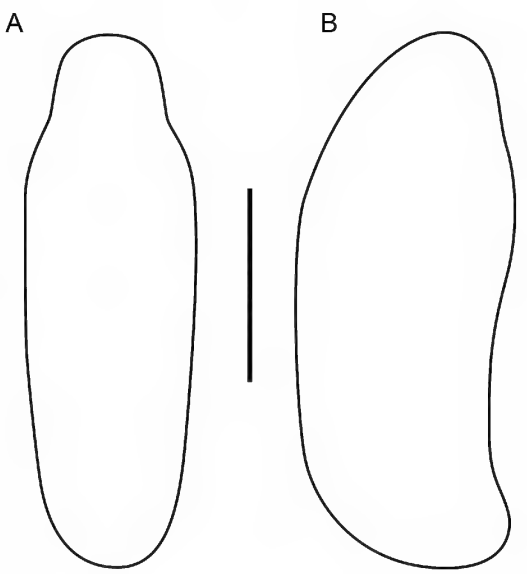


FIG. 52. Diagrams of egg of *Melipona* (*Eomelipona*) *bicolor* Lepeletier, in **A,** dorsal and **B,** side views (scale bar = 1 mm).

Among all meliponine taxa examined, only in *T.* (*Frieseomelitta*) *varia* and *T.* (*Duckeola*) *ghilianii* are the anterior and posterior points of contact along the mandibular hinge between the head capsule and the mandibular surface heavily sclerotized and thus darkly pigmented (fig. 43A). This suggests that the broad, scoop-shaped mandibles require strong hinging to the head capsule. The functioning of this distinctive mandibular attachment to the head needs further investigation. Interesting is the fact that while the relative head size to body size of *T.* (*Frieseomelitta*) *varia* is small (fig. 21) that of *T.* (*Duckeola*) *ghilianii* is much larger (fig. 20).

Material Studied: 9 postdefecating larvae, all with mandibular features as described above: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.A.B. Almeida and D.A. Alves.

Predefecating Larva of *Trigona* (*Geotrigona*) *mombuca* Smith

Figure 44

The two available specimens of this species lacked internally developing pupal features. Therefore their body form was not comparable with that of a fully developed last larval instar and consequently is not illustrated. However, it bore projecting paired dorsal tubercles on the caudal annulets of the thoracic segments and abdominal segments 1-8 that diminished in size toward the posterior end of the body. Furthermore, the dorsal body surface was strongly spiculate, as is characteristic of larval Meliponini. The head capsule was fully developed and bore strongly tapering mandibles with a narrow, pigmented, parallel-sided apex

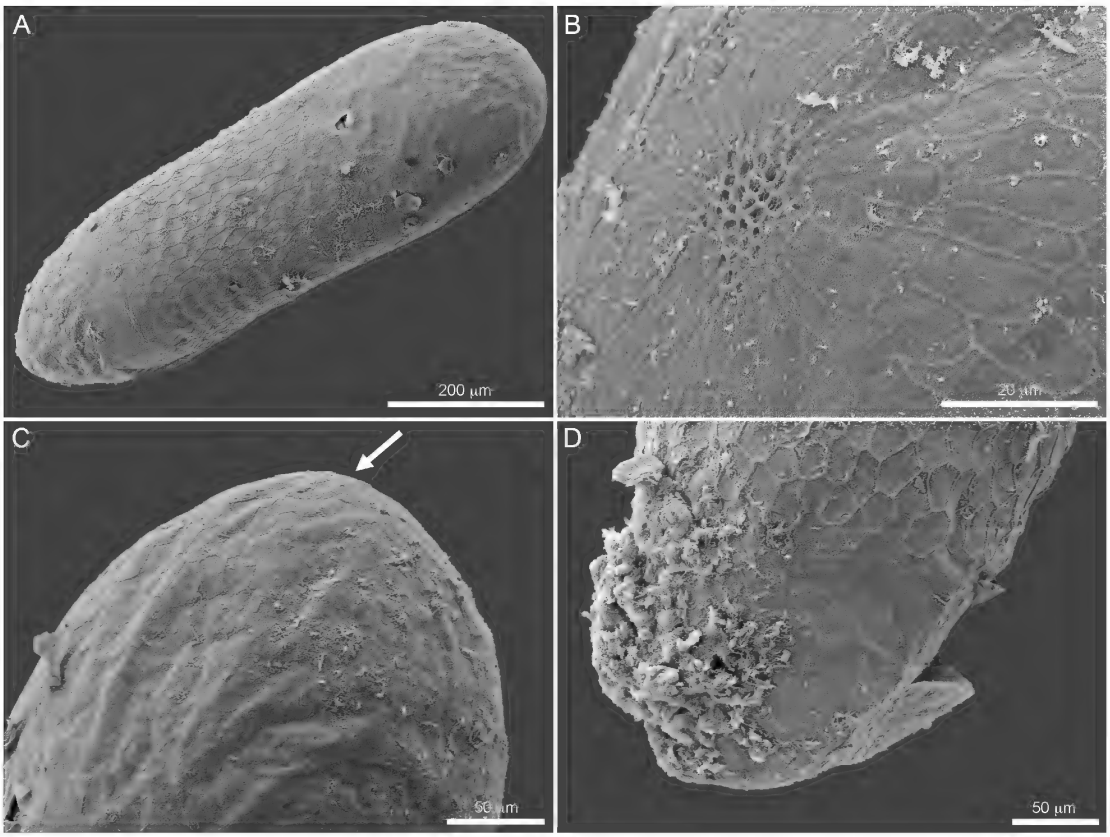


FIG. 53. SEM micrographs of egg of *Austroplebeia australis* (Friese) showing **A,** shape and overall chorionic patterning (anterior end upper right). **B.** Micropyle at extreme apex. **C.** Anterior end close-up showing position of micropyle (arrow) and weak chorionic patterning fading in area of maximum egg width. **D.** Posterior end of egg lacking chorionic patterning.

that seemed to end bluntly, surrounded by fine, sharp spines. On SEM examination, however, the extreme apex appeared to expand into a somewhat spoon-shaped configuration (fig. 44).

Material Studied: 2 early stage last larval instars: Brazil, São Paulo, Ribeirão Preto. 12-XII-1988. J.M.F. Camargo (Nest 480c).

Postdefecating Larva of *Trigona* (*Heterotrigona*) *carbonaria* (Smith)

Figures 19, 40

Rozen and Smith (2019: figs. 11, 13–16) recently published the description of the postdefecating larvae of *Tetragonula sapiens* (Cockerell) from British New Guinea with SEM micrographs of their extremely slender, sharply pointed mandibles and multipronged labral tubercles, similar to those of figure 32B. The following is the description of *Tetragonula carbonaria* (Smith).

Description: **Head:** Size moderately small relative to body size (fig. 19); vertex not bilobed in frontal view. Antennal papilla tuberculate. Posterior margin of head capsule, dorsal view, bend-

ing forward only slightly at midline. Apical surface of labrum (fig. 40A, B) with dense mass of multipronged spicules and sensilla with hemispherical bases.

Mandible (fig. 40B) tapering to vary narrow parallel-sided, nonpigmented, pointed apex bearing long, sharp-pointed spines mostly along dorsal apical edge.

Body: Dorsal integument of thorax spiculate as described for tribe; paired dorsolateral tubercles of three thoracic segments and first abdominal segment small but clearly pointed; those of following abdominal segment scarcely expressed (fig. 19). Spiracles moderately large relative to body size, as described for tribe.

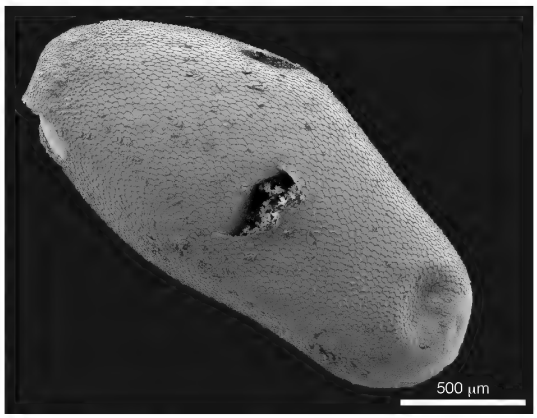


FIG. 54. SEM micrograph of egg of *Melipona* (*Eomelipona*) *marginata* Lepeletier approximate dorsal view.

Material Studied: 10+ mature larvae. Australia, New South Wales, Yama. 03-XII-2019. E.A.B. Almeida.

Postdefecating Larva of *Trigona* (*Tetragona*) *clavipes* (Fabricius)

Figures 22, 45A, B

Description: **Head:** Size small relative to body size (fig. 22); vertex vaguely bilobed in frontal view (fig. 45A).

Mandible apically pigmented and, as seen in outer view (fig. 45B), slender, tapering evenly from base to narrowly rounded apex; apical concavity narrow; dorsal and apical edges of concavity with numerous long, slender, sharp spines as seen in outer (fig. 45B) or inner view; cuspal area not strongly produced, without spicules.

Body: Anterior body segments with distinct but small, unpigmented, projecting, paired dorsolateral tubercles; those of abdomen reducing in size toward posterior end of abdomen.

Material Studied: 5 postdefecating larvae: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

Postdefecating Larva of *Trigona* (*Tetragona*) *nordestina* (Camargo)

Figures 23, 46A–D

Description: **Head:** Size moderately small relative to body size (fig. 23); head not bilobed in frontal view (fig. 46A). Vertex in lateral view curving to meet narrow rim of head capsule. Antenna dome shaped (fig. 46C). Apical surface of labrum bearing extensive patch of elongate, multipronged spicules (fig. 46A, D) that are sufficiently large to be detectible with compound microscope.

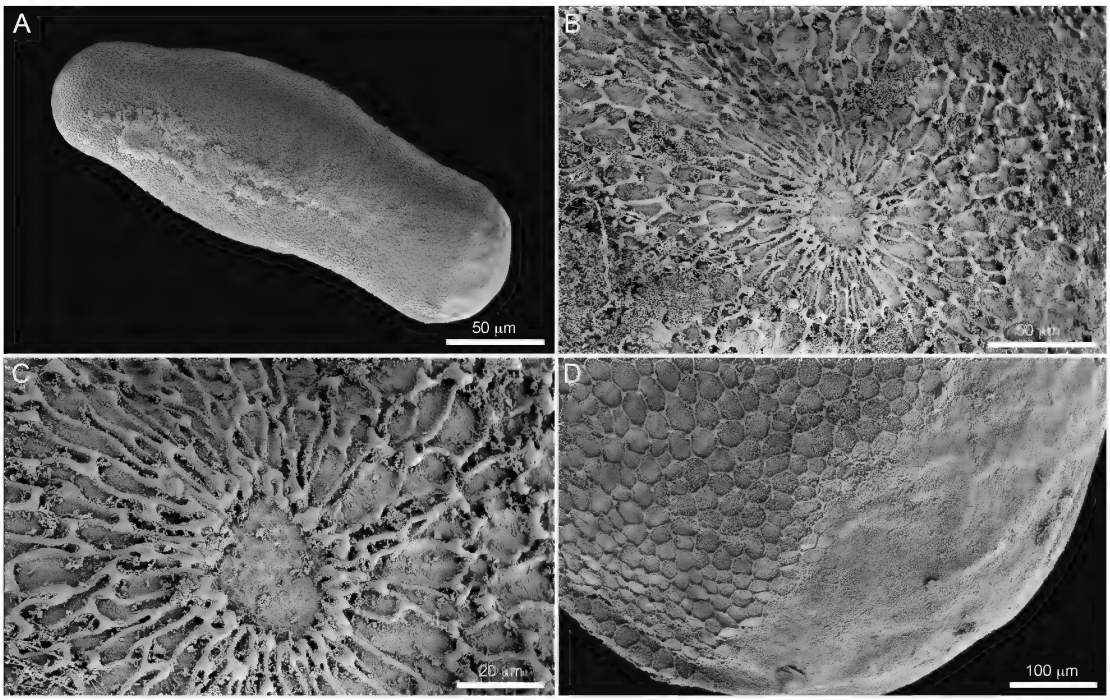


FIG. 55. SEM micrographs of egg of *Melipona* (*Melipona*) *quadrifasciata* Lepeletier. **A.** Entire egg, side view, anterior end upper left. **B.** Anterior end showing micropyle. **C.** Close-up of micropyle. **D.** Extreme posterior end without reticulations.

Mandibular apex pigmented; mandible seen in outer (fig. 46B, D) or inner view extremely slender, narrowing evenly from base to very narrow subapical region and then expanding somewhat at apex; apical concavity obviously narrow but expanding at apex to form a small spoon-like apical concavity bearing fringelike adorally directed spines.

Body: Thoracic and most abdominal segments with clearly defined but small, mostly non-sclerotized paired elevations that are too low to be designated tubercles although their position is at the apex on slightly elevated, transversely oblong stained surfaces of caudal annulet.

Material Studied: 2 postdefecating larvae: Brazil, Piauí, Bom Jesus (Uruçuí-Uma). 4–10-III-1984. J.M.F. Camargo (Nest 551c). 5 postdefecating larvae: Brazil, Piauí, 30 km NW Bom Jesus. 28-I-1993. J.M.F. Camargo (Nest 32c).

Post- and Predefecating Larvae of *Trigona* (*Tetragonisca*) *angustula* Latreille

The predefecating larva of this species was described by Rozen et al. (2019b: figs. 49–52) based on specimens from Panama. The reader is referred to that treatment concerning the tentorium and details of the form of the predefecating mature larva.

Description: **Head:** Size small relative to body size (Rozen et al., 2019b: fig. 50); vertex of head capsule slightly bilobed in frontal view (Rozen et al., 2019b: fig. 51).

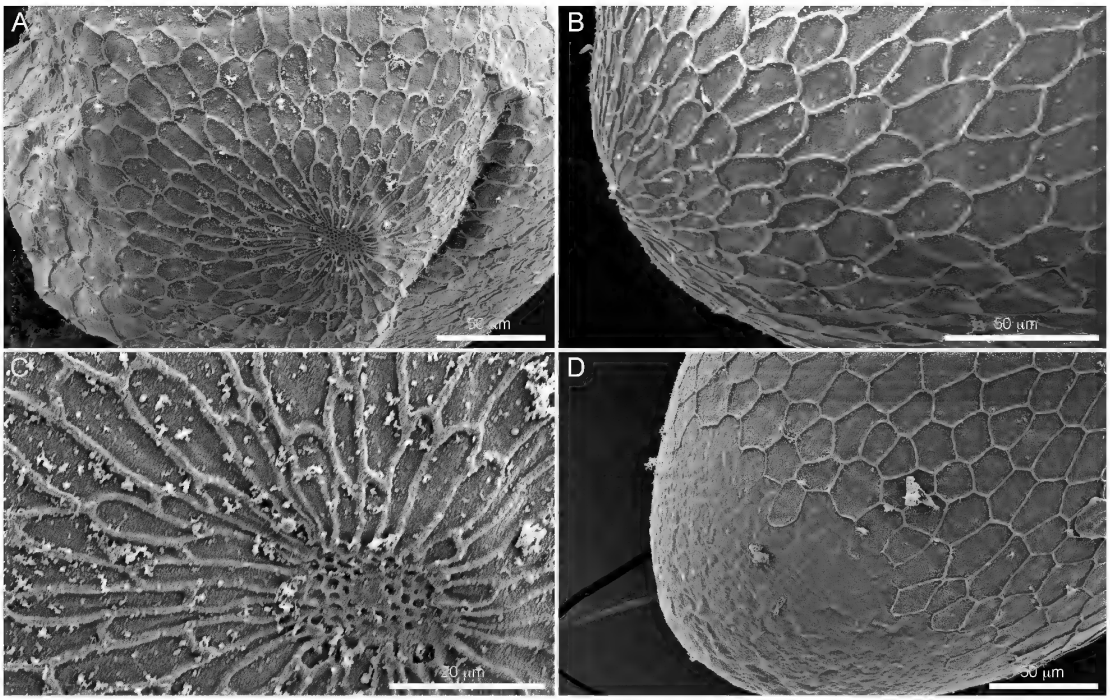


FIG. 56. SEM micrographs of egg of *Nannotrigona melanocera* (Schwarz). **A.** Front end showing geometric patterning of ridges around micropyle. **B.** Side view of same. **C.** Close-up of micropyle. **D.** Lateral view of posterior end of egg showing loss of hexagonal surface patterning.

Mandibular apex at most faintly pigmented; mandible (Rozen et al., 2019b: figs. 49, 51) moderately slender, tapering evenly from base to narrow apex; apical concavity narrow; dorsal edge of apical concavity with numerous long, slender, sharp spines (Rozen et al., 2019b: fig. 49); ventral edge also with sharp spines but fewer in number and extending only to base of concavity; mandibular extreme apex hooked, turning adorally, with numerous fine, sharp spines (Rozen et al., 2019b: fig. 49).

Body: Dorsal integument of body from posterior margin of head toward posterior end moderately covered with fine spicules that become sparser posteriorly; anterior body segments with distinct but small projecting paired dorsolateral tubercles, of which thoracic ones occasionally faintly pigmented; paired dorsolateral tubercles of thorax and anterior abdominal segments spiculate; spiculation of posterior body segments gradually become less distinct. Spiracles moderately large relative to body size; peritreme distinct; atrium shallow; atrial wall more or less concentrically ringed; primary tracheal opening a simple rim, smaller than atrial opening; subatrium moderately short; flexure collapsed into single, long narrow tube.

Material Studied: 10+ postdefecating larvae: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.A.B. Almeida and D.A. Alves. 4+ predefecating larvae: Panama, Colón Prov., 4 km SE of Sabanitas, Roubik Preserve. March 2018. D. Roubik.

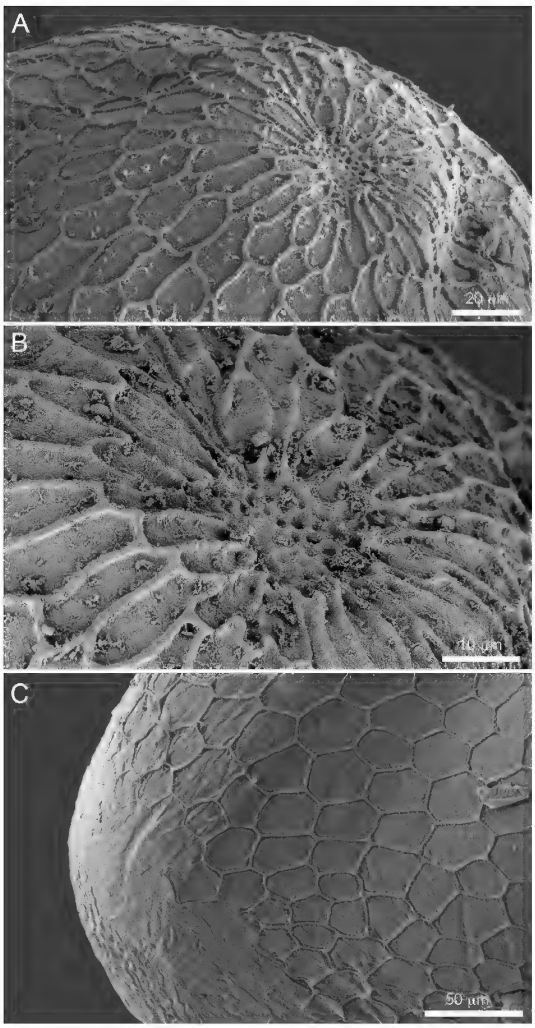


FIG. 57. SEM micrographs of egg of *Plebeia* (*Plebeia*) "nan2." **A.** Front end of egg showing micropyle and patterning of geometric ridges. **B.** Close-up of micropyle. **C.** Loss of geometric patterning at extreme posterior end of egg.

Trigona (*Trigona*) *corvina* Cockerell

The mature larva of *Trigona corvina* Cockerell was briefly described by Michener (1953) who also illustrated its mandible and spiracle.

Trigona (*Trigona*) *crassipes* (Fabricius)

Figures 24, 47, 48A–C

Description: **Head:** Size small relative to body size (fig. 24); vertex faintly bilobed in frontal view.

Mandibular apex nearly unpigmented in that faint tinting detected with microscope only on cleared specimen (except this specimen with atypical apical brown spot); mandible in outer view (figs. 47, 48A–C) slender, tapering from base to apex; combined sides of apex and extreme apex forming shallow apical concavity; entire dorsal edge of apex densely fringed with row of tapering spines extending to narrow apex and around hooked-shape extreme apex for short distance along lower apical edge.

Body: Anterior body segments with distinct but very small, projecting, paired dorsolateral tubercles; those of three thoracic segments often faintly pigmented; paired abdominal tubercles weakly expressed, becoming increasingly faint toward rear of abdomen.

Material Studied: 11 postdefecating larvae in their cocoons: Brazil, Amazonas, Ipixuna, Purus, 42 km from Tapauá. 22-I-1986. J.M.F. Camargo (Nest 339c).

Remarks: The cocoon fabric is sufficiently transparent that lack of mandibular pigmentation was obvious even before specimens were removed from their cocoons.

Trigona (*Trigona*) *hypogea* Silvestri

Figure 25

Except for its smaller size (table 1; compare figs. 24 and 25, diagrammed to the same scale) and for larger head size relative to body size, characterized as moderate, the larva of this carrion-collecting species is similar to that of *T. crassipes,* described above.

Material Studied: 15 postdefecating larvae: Brazil, Amazonas, Bacururu, rio Juruá. 31-VII-1993. 1J.M.F. Camargo (Nest 545c).

Trigona (Trigona) *pallens* (Fabricius)

Figure 26

With nearly the same body size of the mature larva of *T. crassipes*, the larva of *T. pallens* differs little from that of *T. crassipes* or, except for size, from that of *T. hypogea*. However, paired dorsolateral tubercles of *T. hypogea* are somewhat smaller than those of the others.

Material Studied: 8 postdefecating larvae: Brazil, Amazonas, Tefé. 28-I-1977. J.M.F. Camargo (Nest 108c).

Trigona (*Trigona*) *spinipes* (Fabricius)

Figures 27, 49A, B

The mature robust larva of *T. spinipes* is nearly identical to that of *Trigona crassipes*, described above. The mandible is figured (fig. 49A, B).

Material Studied: 6 mature larvae: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

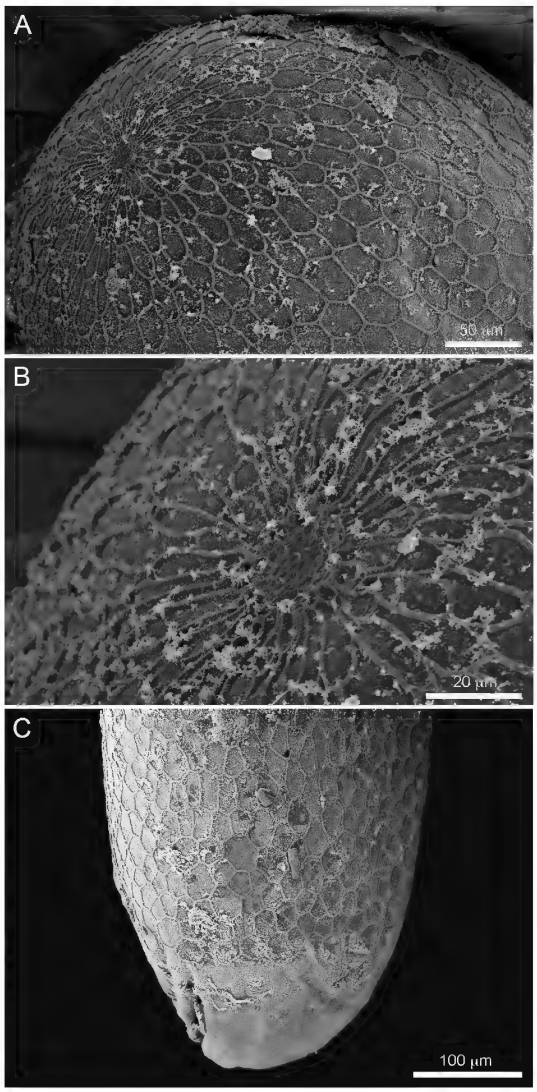


FIG. 58. SEM micrographs of egg of *Trigona* (*Frieseomelitta*) *varia* (Lepeletier). **A.** Front end. **B.** Close-up of micropyle. **C.** Posterior end showing loss of surface patterning.

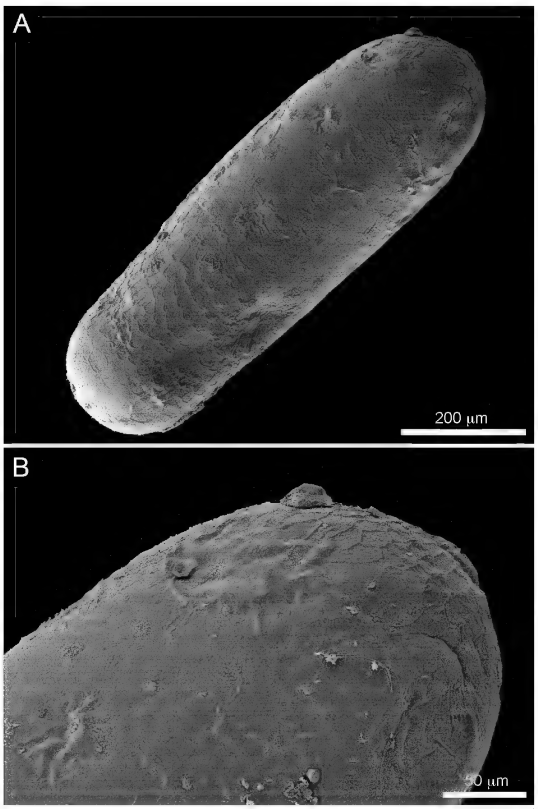


FIG. 59. SEM micrograph of egg of *Trigona* (*Trigona*) *spinipes* (Fabricius). **A.** Entire egg, lateral view, revealing smooth posterior end and chorionic patterning elsewhere. **B.** Anterior end showing weak chorionic ridges and with arrow pointing to micropyle.

Postdefecating Larvae of *Trigonisca*

Trigonisca longicornis (Friese)

Figures 28, 50A–C

Description: **Head:** Size moderately small relative to body size (fig. 28). Antennal papilla a large, low, nontuberculate dome (fig. 50A). Labrum rather narrow with apical width clearly less than intraantennal distance.

Mandibular apex scarcely pigmented but distinctly sclerotized; mandible seen in outer (fig. 49B, D) or inner view apically extremely slender, fingerlike, narrowing evenly from base to apex; apex elongate, tapering to sharply pointed tip and bending adorally; dorsal edge of mandible with linear row of perhaps five elongate spines.

Labiomaxillary region as described for tribe.

Body: Dorsal integumental spiculation about as described for tribe. Thoracic and most abdominal segments with low, paired dorsolateral elevations on caudal annulets. Dorsolateral paired elevation of three thoracic segments and of first abdominal segment each with small unpigmented tubercle; tubercles becoming progressive less distinct on abdominal segment to become nearly unrecognizable beyond abdominal segment 5 although abdominal segment 8 with paired, nonsclerotized elevations on slightly elevated, transversely oblong stained surfaces of caudal annulet; on abdominal segments 9 and 10 each pair united medially forming single, slightly elevated bar of caudal annulet. Spiracles as described for tribe.

Material Studied: 4 postdefecating larvae: Brazil, Goiás, Aragarças. 10–24-I-1971. J.M.F. Camargo (Nest 09c).

Trigonisca mepecheu Engel and Gonzalez

Figure 29

The postdefecating larva and egg of this species from Colombia were recently described by Rozen in Engel et al. (2019) with detailed information on its nesting biology and ethnographic data. Except for size, there is a close similarity between postdefecating larva of *Trigonisca mepecheu* and

the mature larva of *Trigonisca longicornis* (fig. 28). Both have large, domelike (i.e., not tuberculate) antennal papillae and mandibles that are identical in that they taper to a sharply pointed, curved apex, as illustrated for *T. mepecheu* in Engel et al., 2019: figs. 11, 12D–G).

Trigonisca muelleri (Friese)

Figures 30, 51A–E

The mature larva of this species was originally described and illustrated by Lucas de Oliveira (1970: figs. 1–3) as *Hypotrigona* (*Leurotrigona*) *muelleri* (Friese). One of the two larvae cleared for the current study could not be prepared for SEM examination, but the other, possibly a fourth instar (fig. 30), demonstrates well characteristic features of the other larval *Trigonisca*, i.e., the large dome-shaped antennae (fig. 51E) and mandibles that strongly taper and bend apically toward the narrow, sharply pointed apex with dorsal row of spines (fig. 51A, B, D).

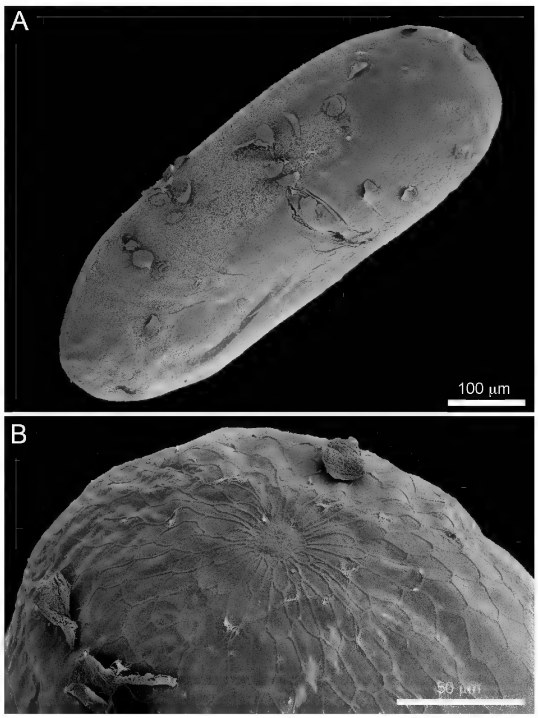


FIG. 60. SEM micrographs of egg of *Trigonisca* (*Trigonisca*) *muelleri* (Friese). **A.** Entire egg, upper end toward right. **B.** Anterior end with micropyle.

Material Studied: 3 post- and 3 predefecating larvae: Brazil, São Paulo, Jaguariuna. 7-I-2020. E.A.B. Almeida.

CONCLUDING REMARKS

Others working on groups within the Meliponini have long recognized that larval Meliponini are remarkably uniform, exhibiting little anatomical variation. However, for the first time it is recognized the mature larvae of *Trigona* (*Duckeola*) *ghilianii* and *Trigona* (*Frieseomelitta*) *varia*, while clearly belonging to this tribe, exhibit striking features that suggest a close relationship with one another and that separate them from other tribal members. One phylogenetic hypothesis proposed for Meliponini based on 34 characters from adult anatomy and nesting biology (Camargo and Pedro, 2003) and one DNA-based hypothesis (Rasmussen and Cameron, 2010) suggested that *T.* (*Duckeola*) and *T.* (*Frieseomelitta*) are close relatives. These meliponine taxa uniquely share: (1) the greatly enlarged and otherwise modified mandible armed with its uniquely swollen, darkly pigmented, anterior and posterior points of articulation with the head capsule, and (2) the spiculate front of the head between the antennae and above the clypeus. The enlarged mandible and its strengthened base almost certainly indi-

cate that the two taxa use their mandibles for one or more special, currently unknown tasks not required of larvae of other stingless bees. In addition, examination of the outer surface of their broad mandibles reveals a dense, nearly total covering of multipronged spicules (figs.41b, 43A), suggesting that in action they function to reduce surface tension created when they rapidly plow through sticky provisions. Mandibles of all other known meliponine taxa (with at most only a few spicules) taper strongly from a thick base to an extremely narrow apex. Thus, the mandibular apex provides far less resistance as the mandible opens and closes during feeding on the sticky provisions. Although Michener (2013) thought that *Tetragonisca* might share a close relationship with *Frieseomelitta* and *Duckeola*, larval anatomy of *Tetragonisca angustula* provides no supporting evidence.

Multipronged spicules found elsewhere serve other purposes. As pointed out by Rozen and Smith (2019) multipronged spicules on the apical labral surface of meliponine bee larvae assist in food ingestion by pushing or raking provisions toward the esophagus. Also, there is a possibility that when multipronged spicules are aligned side by side, their aligned bases may reinforce integument that is somewhat flexible, so that it resists bending from one direction but encourages it from another direction, as is nicely demonstrated in figure 50C, a highly magnified view of the outer surface of the right maxilla of the immature larva of *Trigonisca muelleri* where the rings of spicules allow the maxilla to project forward but constrain the maxilla in moving dorsally, ventrally, or side to side.

ADDENDUM: DESCRIPTIONS OF EGGS OF MELIPONINI

Figures 52–59

As indicated in Rozen et al. (2019b), the known egg shape of the Meliponini appears somewhat asymmetrical around the long axis of the egg. This asymmetry is probably characteristic for the tribe but too slight to warrant description in both front/back and lateral views. In the following descriptions, eggs are assumed to be oriented with anterior (micropyle) end higher than the posterior end. The symmetrical outline, illustrated in most cases, is assumed to be the front or back. The symmetrical outline and the asymmetrical outline at 45° from the symmetrical one are shown here for *Melipona bicolor* (fig. 52A, B). This asymmetry in meliponine eggs was also noted for *Melipona beecheii* Bennet, *M. panamica* Cockerell, *Scaptotrigona pectoralis* (Dalla Torre), and *Tetragonisca angustula* Latreille (Rozen, et al., 2014).

Measurements of all species except for *Trigonisca muelleri* were taken from specimens that had not been critical-point dried. Noteworthy is that egg measurements of *Nannotrigona testaceicornis* were recorded both before and also after critical-point drying and revealed, not surprisingly, a loss of dimensions due to this treatment, as reported below. All other eggs were measured while in preservative and without critical-point drying.

Table 2 lists the taxa the eggs of which have previously been examined with an SEM.

***Melipona* (*Eomelipona*) *bicolor* Lepeletier:** Length 2.9 mm, maximum width 0.9 mm more or less frontal view 1.2 mm lateral view short distance above midbody ($N = 1$) (fig.

TABLE 1. List of species of Meliponini whose mature larvae either are described here or were described earlier, as referenced parenthetically. This list includes representatives of 26 of approximately 47 genera and subgenera included in the tribe. Measurements demonstrate comparative lengths of mature postdefecating larvae based on inter-spiracular distances (as calculated in Rozen and Smith, 2019), modified as indicated in Methods (above). However, measurements preceded by "est." (for estimated) were derived by taking a series of measurement along one side of those larvae all of whose spiracles could not be clearly identified. Genera arranged alphabetically. **LL,** larval length, is shown in mm.

Taxon	LL
Austroplebeia australis (Friese)	6.8
Cephalotrigona capitata (Smith)	11.2
Lestrimelitta ehrhardti (Friese) (Lucas de Oliveira, 1968)	
Lestrimelitta glabrata Camargo and Moure	9.3
Lestrimelitta limao (Smith) (Lucas de Oliveira, 1968)	8.6
Melipona (*Eomelipona*) *bicolor bicolor* Lepeletier	9.2
Melipona (*Eomelipona*) *bicolor schencki* Gribodo (as *Melipona nigra schencki* Gribodo in Lucas de Oliveira, 1960)	–
Melipona (*Eomelipona*) *marginata* Lepeletier (Michener, 1953)	–
Melipona (*Melipona*) *quadrifasciata* Lepeletier (Michener, 1953)	–
Melipona (*Melipona*) *variegatipes* Gribodo (Michener, 1953)	–
Melipona (*Michmelia*) *fallax* Camargo and Pedro (Rozen et al., 2019b; Rozen and Smith, 2019)	18.3
Melipona (*Michmelia*) *trinitatis* Cockerell (Rozen et al., 2019b)	16.2
Nannotrigona melanocera (Schwarz)	6.8
Nannotrigona testaceicornis (Lepeletier)	5.9
Nogueirapis mirandula (Cockerell) (Rozen et al., 2019b)	6.2
Oxytrigona tataira (Smith)	7.4
Paratrigona impunctata (Ducke)	7.8
Paratrigona lineatifrons (Schwarz)	6.3
Partamona cupira (Smith)	7.5
Partamona peckolti (Friese) (as *Trigona* (*Partamona*) *cupira* Smith in Michener, 1953)	–
Partamona musarum (Cockerell) (Rozen et al., 2019b)	8.8
Plebeia (*Plebeia*) *droryana* (Friese) (Lucas de Oliveira, 1965)	5.9
Plebeia (*Plebeia*) "nan1" (Rozen et al., 2019a)	6.4
Plebeia (*Plebeia*) "nan2"	4.2
Plebeia (*Plebeia*) *schrottkyi* (Friese)	4.8
Plebeia (*Scaura*) *latitarsis* (Friese)	est. 5.8
Scaptotrigona bipunctata (Lepeletier)	9.1
Scaptotrigona depilis (Moure)	7.4
Scaptotrigona polysticta Moure	–
Scaptotrigona postica (Latreille) (Lucas de Oliveira, 1960)	–
Tetragonula carbonaria (Smith)	5.9
Tetragonula sapiens (Cockerell) (Rozen and Smith, 2019)	5.9
Trigona (*Duckeola*) *ghilianii* (Spinola)	12.9
Trigona (*Frieseomelitta*) *varia* (Lepeletier)	7.8
Trigona (*Geotrigona*) *mombuca* (Smith)	est. 7.7
Trigona (*Tetragona*) *clavipes* (Fabricius)	8.7
Trigona (*Tetragona*) *nordestina* (Camargo)	9.0
Trigona (*Tetragonisca*) *angustula* Latreille (Rozen et al., 2019b)	6.8
Trigona (*Trigona*) *corvina* Cockerell (Michener, 1953)	–
Trigona (*Trigona*) *crassipes* (Fabricius)	10.6
Trigona (*Trigona*) *hypogea* Silvestri	7.5
Trigona (*Trigona*) *pallens* (Fabricius)	8.7
Trigona (*Trigona*) *spinipes* (Fabricius)	9.1
Trigonisca longicornis (Friese)	5.8
Trigonisca mepecheu Engel and Gonzalez (Engel et al., 2019) (as *Trigonisca* "nan1" in Rozen and Smith, 2019)	3.8
Trigonisca muelleri (Friese) (Lucas de Oliveira, 1968)	est. 4.4

TABLE 2. List of additional meliponine taxa whose eggs have previously been examined and described by SEM.

Melipona (*Melikerria*) beecheii Bennett	Rozen et al., 2019b
Melipona (*Michmelia*) *panamica* Cockerell	Rozen et al., 2019b
Plebeia (*Plebeia*) "nan1"	Rozen et al., 2019a
Scaptotrigona pectoralis (Dalla Torre)	Rozen et al., 2019b
Tetragonisca angustula (Latreille)	Rozen et al., 2019b

52A, B). Exaggerated shape especially in lateral view (fig. 52B) possibly created by developing embryo. Micropyle at anterior end; chorion with elevated network of hexagons and other geometric figures except at posterior end where smooth.

Material Studied: 1 egg: Brazil, São Paulo, Jaguariuna. 27-I-2020. E.A.B. Almeida.

***Melipona* (*Eomelipona*) *marginata* Lepeletier:** Length 2.48 mm; maximum width short distance above midbody 1.18 mm (N = 1); shape (fig. 54) of egg almost certainly indicating it to be symmetrical when in full front/back view and asymmetrical in side view. Micropyle dorsal at anterior end, well surrounded by chorionic ridges though weakly visible; posterior end smooth, without reticulations.

Material Studied: 1 egg: Brazil, São Paulo, Jaguariuna. 27-I-2020. E.A.B. Almeida.

***Melipona* (*Melipona*) *quadrifasciata* (Lepeletier):** Length 2.45–2.5 mm, maximum diameter 1.0–1.1 mm (N = 4) short distance above midbody (fig. 55A). Nearly hemispherical apical end (fig. 55A) expanding evenly to maximum diameter and then tapering to posterior end just before which it expands and then terminates. Micropyle (fig. 55B, C) at anterior end; chorion with elevated network of hexagons and other geometric figures except at posterior expansion where smooth (fig. 55D).

Material Studied: 4 eggs: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.B.A. Almeida and D.A. Alves.

***Melipona* (*Michmelia*) *panamica* Cockerell:** Both eggs were badly damaged in that the posterior end of one was completely missing and the posterior end of the other egg was an empty fragment of the chorion attached to the front end. However, enough of both eggs remained to indicate that the general shape is similar to that of other known *Melipona*. Length ca 2.5 mm; maximum diameter 1.0–1.1 mm at ca 1 mm from anterior end (N = 2). Slightly projecting micropyle centered at anterior end; chorion with elevated design of hexagons that at anterior end morph into more slender geometric patterns pointing toward micropyle; chorion at posterior end smooth, lacking geometric pattern.

Material Studied: 2 eggs: Panama, Colón Prov., 4 km SE of Sabanitas, Roubik Preserve. March 2018. D. Roubik.

***Nannotrigona testaceicornis* (Lepeletier):** Length 1.0–1.075 mm, maximum diameter 0.4 mm (N = 3) (fig. 56). Measured again after critical-point drying: length 0.875–0.9 mm; maximum diameter 0.325–04.4 mm. Slightly longer than egg of *Plebeia droryana* but also with anterior end broadest: egg tapering posteriorly somewhat more than in *P. droryana*; micropyle centered on anterior end (fig. 56A, B, C); chorion surface reticulate as in *P. droryana*; posterior end smooth (fig. 56D).

Material Studied: 3 eggs: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

***Plebeia* (*Plebeia*) *droryana* (Friese):** Length 0.88 mm (N = 1), maximum diameter 0.4 mm; anterior end, tapering slightly posteriorly with anterior end somewhat blunter than posterior end. Micropyle (fig. 57A, B) at anterior end; chorion with elevated network of hexagons and other similar geometric figures that elongate around micropyle; these geometric figures ceasing abruptly near posterior end, so that extreme posterior end of chorion smooth (fig. 57C).

Material Studied: 1 egg: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

***Plebeia* (*Plebeia*) "nan2":** Average length 1.03 mm (N = 10), maximum diameter 0.325 mm (N = 1), poorly preserved; approximately parallel sided, anterior end with micropyle perhaps slightly blunter than posterior end; chorion with strongly developed pattern of geometric figures except for posterior end where smooth.

Material Studied: 10 eggs: Ecuador, Francisco de Orellana Prov., Yasuni Scientific Field Station, Pontifica Universidad Católica del Ecuador, -0.8659, -76.3953. Dec. 10, 2019. D. Roubik.

***Trigona* (*Frieseomelitta*) *varia* (Lepeletier):** Length 1.25 mm, maximum diameter 0.43 mm (N = 5) at anterior end (fig. 58) tapering toward more narrowly rounded posterior end (fig. 58C). Micropyle centered on anterior end (figs. 58A, B); chorion extensively covered with elevated network of hexagons and other geometric figures, extreme posterior end smooth (fig. 58C), without such patterning.

Material Studied: 5 eggs: Brazil, São Paulo, Piracicaba. 13-VII-2019. E.A.B. Almeida and D.A.I Alves. 6+ eggs: Brazil, São Paulo, Jaguariuna. 21-I-2020. E.A.B. Almeida.

***Trigona* (*Trigona*) *spinipes* (Fabricius):** Length 1.05 mm; maximum width 0.33 mm about ¼ distance from anterior end where reticulate chorion pattern weakly expressed (fig. 59A). Micropyle at extreme apex (fig. 59B: arrow). Below maximum diameter, sides of egg with more obvious reticulations converging slightly until reaching rounded posterior end with smooth chorion.

Material Studied: 6 eggs: Brazil, São Paulo, Jaguariuna. 21-VII-2019. C. Menezes.

***Trigonisca muelleri* (Friese):** Length 0.63–0.7 mm, maximum diameter 0.234–0.27 (N = 4) at about ¼ distance from anterior end, slightly tapering posteriad from there, so as to almost seem parallel sided until curving to posterior end, which is slightly more pointed than nearly semicircular anterior end (fig. 60A, B); long axis slightly curved; micropyle (fig. 60B) approximately centered on anterior with typical array of elevated reticulate chorion ridges converging toward central disc of micropylar openings; weakly elevated ridges probably resulting from deterioration after preservation; posterior end lacking reticular patterning.

Material Studied: 4 eggs: Brazil, São Paulo, Jaguariuna. 27-I-2020. E.A.B. Almeida.

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